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LOOKING FORWARD

SOIL SCIENCE is now in its twenty-fifth year, and will soon have completed its fiftieth volume. Its success as a medium for the publication of technical papers dealing with the science of the soil, and closely related subjects, has long since been assured. The journal enjoys a wide circulation, both in America and abroad, and is receiving manuscripts from all over the world.

From its inception, the columns of SOIL SCIENCE have been open to all workers in the field of soils, and many of them have taken advantage of this privilege. Over the years, the number of papers submitted has markedly increased, and their quality has greatly improved. The scope of soil research has now reached such proportions that additional assistance is required in dealing with the manuscripts which are sent in for consideration.

Accordingly, it seemed advisable to increase the number of consulting editors, and this has been done. The men whose names appear on the editorial list for the first time in this issue were carefully chosen by the old editorial staff. They are outstanding scientists in their respective fields of research. Other equally capable men could and would have been added to the list except for the fact that a very large editorial board is unwieldy.

Manuscripts will now receive more careful attention than was previously possible. Papers will be read and accepted within a short time after their receipt, or returned to the authors with reasons for their rejection. If accepted, any suggestions of the editorial readers which will serve to improve the presentation of the material will be forwarded to the authors for their consideration.

The editor-in-chief is not unmindful of the honor which has been bestowed upon him in having been chosen to direct this work. With this honor comes the obligation to maintain the high standards set by Dr. Jacob G. Lipman, the founder of SOIL SCIENCE, and its guiding spirit over a quarter of a century. That is our aim.

FIRMAN E. BEAR.

DETERMINATION OF SOIL STRUCTURE BY MICROSCOPICAL INVESTIGATION

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It is important to know what changes are brought about in the structure of soil by the operations of cultivation. Various methods have been proposed for estimating the size and form of soil particles and of the pore spaces enclosed by them². As none of the methods hitherto available is visual, however, an adaptation of Kubiena's thin-section method³ has been worked out for measuring, with the aid of a microscope, the actual pore spaces in a specimen of substantially undisturbed soil.

This new method makes possible a literal insight into soil "structure," in the sense of the word as used by Russell⁴. It also enables subjective impressions of photographic or other records to be made of the structure of a single sample or profile of soil and affords a quantitative expression of a linear function of the pore space, so that the pore spaces of different samples can be expressed in terms of a ratio.

DESCRIPTION OF THE METHOD

A special mode of sampling is used for this modification of Kubiena's technic. Sampling tubes are made from steel boiler tube having an internal diameter of 4.7 cm. and cut into 3.8-cm. lengths, each of which is beveled into a sharp edge at one end. The sampling tube is driven into the soil at right angles to the soil exposure which is to be sampled. When the top of the ring is level with the exposed surface, the ring and its contents are withdrawn by passing a knife around the outside. The core is then trimmed level, without being removed from the ring, and its ends are covered with pieces of tin plate held in position by a rubber band. Orientation and reference marks are made on the ring immediately after withdrawal. For transporting a series

¹ I am indebted to J. Hudig for providing me with laboratory facilities and for allowing me to use as experimental material the soils treated with the humic material. These treatments were performed in connection with an investigation by Professors Hudig and Van Reesema, of which the results are not yet published. To the Imperial Bureau of Soil Science, Rothamsted Experimental Station, I am indebted for the revision of this paper.

² Keen, B. A. 1931 *The Physical Properties of the Soil*. Rothamsted Monographs on Agricultural Science. Longmans, London.

³ Kubiena, W. L. 1938 *Micropedology*. Collegiate Press, Ames, Iowa.

⁴ Russell, E. W. 1938 *The structure of soil*. Imp. Bur. Soil Sci. Tech. Commun. 37.

of samples a box containing a number of square or round compartments, each just large enough to allow a ring to fit in loosely, is convenient. The divisions should be not more than 3 cm. high, so that the sampling rings can be easily withdrawn before and after use.

If the sample consists of a noncoherent soil, such as a sand or a crumbly clay, it should first be air-dried in the laboratory, within its ring, and then be immersed in cellulose lacquer diluted with about an equal volume of acetone. After evaporation of the acetone, the sample may be removed and further manipulated. Otherwise, the preparation of thin sections (about 0.03 mm. thick) is as described by Kubiena.

It is not advisable to make more than one or two sections from any one core. If the section is made from about the middle of the height of the core it may be safely assumed to represent an undisturbed sample of soil in its natural condition. If on removal from its ring a core breaks up, the larger pieces should not be selected for sectioning, as such pieces, being essentially clods, have a poorer structure than the average of the whole sample.

Even without a microscope it is possible to get an idea of the quality of the structure of a soil by looking at a well-made thin section. A section having a poor structure looks like a map of a continent, that is, the solid masses predominate (pl. 1, fig. 5); a section having a good structure, on the other hand, resembles an archipelago, the solid masses being small and dispersed, yet preserving a recognizable entity (pl. 1, fig. 2).

For microscopic measurements of the sections, a Leitz polarizing microscope furnished with objective 3 of N.A. 0.25 and with eyepiece PM 6 \times containing a micrometer scale was used. One division of the micrometer was equal to 16 μ . In polarized light and seen through a gypsum plate, clay appears dark, crystals show interference colors, and the pore spaces, which are filled with the resinous material ("Kollolith") used in Kubiena's preparation, appear violet. Clay and sand can therefore be readily distinguished from pores.

The section is placed on the mechanical stage, and every visible pore and grain wider than 16 μ is measured. Measurements are made only along the direction of travel (without respect to sign), the micrometer scale being adjusted so that it lies along this line. Figure 1 illustrates diagrammatically the appearance of four large crumbs in conjunction with the micrometer scale. The successive crumbs shown are recorded as having "diameters" along the scale of 20, 13, 15, and 4 and 2 divisions (each equal to 16 μ), their dimensions at other angles to the scale being ignored.

The determination of such "diameters" for pore spaces and for crumbs or single grains constitutes the basis of this work. Since it is the ratio of pore space to solid which is especially important in this study, the term "diameter" can be dropped without risk of confusion.

When the edge of the section is reached, the stage is displaced by 1 mm., and a new line, parallel to the previous one, is surveyed. On large sections the survey lines can be spaced at more than 1 mm. It has been found necessary to survey a total length of about 200 mm. for each section in order to ob-

tain reliable averages. For example, in one section over which the survey lines were spaced at 1 mm., the percentages of solids and of pore spaces were 87.35 and 12.65 in the even lines and 86.62 and 13.38 respectively in the odd lines, a total of about 200 mm. being surveyed in each of the two groups.

Calculation of the ratio of pore space to solids, or of the percentage of pore space and/or of solids, can be made either by measuring both pore spaces and solids, the length of line surveyed not being accurately measured, or by measuring accurately the length of line surveyed and the total of either pore spaces *or* solids.

For the tabulations presented in this paper intervals of 10 micrometer divisions ($160\ \mu$) have been adopted: over $16\ \mu$ and less than $160\ \mu$, 160 – $319\ \mu$, 320 – $479\ \mu$, and so on up to $1599\ \mu$, then over $1600\ \mu$.

In a soil of ideal structure the ratio of solids to pore spaces should be 0.5. This ratio is never attained as a result of practical observations by the method here described, for pores smaller than $16\ \mu$ are not measured. The notion that the error introduced by ignoring such small pores in micrometric measurement is negligible receives support from the claim of Sekera⁵ that water contained in the finest pores or elemental cells of soil, like the water film on soil particles, is not available to plants.



FIG. 1. CRUMBS OF THIN SECTIONS AS SEEN IN RELATION TO MICROMETER SCALE

A soil of good structure consists largely of small crumbs, conglomerates of smaller soil particles. The larger are the crumbs, the smaller is the developed inner surface of the soil, which is the area active in plant nutrition. The aim of cultural operations in the field is to enlarge this effective surface area of the soil. As a tentative quantitative expression of the structure of a soil, the following expression has been developed to take these considerations into account:

$$\frac{\text{Percentage of pore space} \times 100}{\text{Percentage of crumbs larger than } 1600\ \mu}$$

The choice of the denominator is arbitrary, as evidence regarding the optimal crumb size for plant growth is not yet available. In the following discussion, a value for the foregoing expression is referred to as the *quotient* for a soil. The larger the quotient is, the better the structure.

APPLICATION OF THE METHOD

The method of microscopical and micrometric examination has been applied to two clay soils of the Netherlands: a marine clay from the Wilhelmina

⁵ Sekera, F. 1933 Die Anpassung der Düngewirtschaft an die Wasserversorgung der Pflanze. *Phosphorsäure* 3: 1–63.

Polder near Goes, and a Rhine clay from Etten. The mechanical analyses of these soils are recorded in table 1.

Soil *in situ* in the field was sampled by the process already described. Samples were taken of untreated soil (controls) and of soil treated with stable humus product⁶ (treated soil). Large samples of both the Goes and the Etten soils were also transported to the Agricultural College at Wageningen, where they were used to form small beds, treated and untreated, in the laboratory garden, in order to observe how application of the humus products affected the soil structure. The micrometric examinations were made at the Wageningen Agricultural College.

Table 2 gives the results of examination of thin sections of the Etten soil *in situ*. From the results, it appears that treatment with the humus product has increased the percentage of small crumbs and has decreased the percentage of crumbs larger than 1600 μ , the latter effect being very striking.

TABLE 1
Mechanical analyses of the Goes and Etten soils
In percentages of oven-dried soil

	GOES	ETTEN
> 50 μ	11.1	27.8
10-50 μ	31.3	11.2
5-10 μ	5.0	10.1
2- 5 μ	4.0	12.2
< 2 μ	34.2	36.1
Organic matter	2.0	2.6
CaCO ₃	12.4
Total.....	100.0	100.0

Table 3 gives for the Etten soil the percentages of solid soil aggregates and of pore spaces, the percentage of crumbs larger than 1600 μ , and the quotients. It will be seen that differences in structure are not well brought out by the percentages of pore spaces or of large crumbs. The quotient, however, does seem to be characteristic of the structure. For example, Nos. 90-3 and 93-4 have similar percentages of pore space, though they are very different in structure. The difference in this case is expressed by the percentages of large crumbs (> 1600 μ). In Nos. 95-3 and 95-7, however, the percentages of large crumbs are similar, but the quotients (respectively 1289 and 741) reflect the difference in structure. These two examples illustrate the need for taking both pore space and percentage of large crumbs into account when considering structure quantitatively. The quotient attempts to do this.

Differences in the structures of samples taken from the surface and at a shallow depth are illustrated by comparing Nos. 93-3 and 95-1-3 with the

⁶ Prepared by Siewertz van Reesema at the Calvé Works, Delft, Holland.

corresponding surface samples. The 3-cm. samples have larger quotients than any of the surface samples. It may be supposed that the smaller quotients of the surface samples represent packing due to heavy rainfall.

TABLE 2
Etten soil: particle or crumb percentages

THIN SECTION NUMBER	TOTAL LENGTH OF CRUMBS AS A PERCENTAGE OF THE OVER-ALL LENGTHS EXAMINED										
	<160 μ	160- 319 μ	320- 479 μ	480- 639 μ	640- 799 μ	800- 959 μ	960- 1119 μ	1120- 1279 μ	1280- 1439 μ	1440- 1599 μ	>1600 μ
Control											
94-1	10.2	9.5	8.8	8.8	9.9	4.6	4.2	3.2	2.5	3.5	22.2
2	6.6	9.1	9.1	7.1	6.1	7.1	3.5	7.1	3.5	3.0	32.3
3	9.4	11.6	5.1	8.7	4.3	7.2	5.8	2.9	2.9	1.4	31.1
4	5.2	9.6	5.9	12.5	6.6	4.4	5.1	4.4	5.2	3.7	31.7
5	11.9	11.9	9.9	4.0	2.0	6.0	4.0	6.0	2.0	37.8
90-1	13.8	16.4	12.3	8.2	5.3	4.4	5.0	2.0	1.7	2.0	8.5
2	10.2	11.8	7.3	8.1	5.7	4.1	5.3	3.2	2.0	1.6	27.2
3	9.4	3.9	6.3	2.4	6.3	2.4	1.6	2.4	4.7	...	37.7
4	5.8	5.4	7.8	8.3	8.3	4.9	3.9	1.0	2.9	2.9	30.6
5	6.3	12.1	8.8	5.8	5.0	2.9	2.5	4.6	1.2	2.1	24.2
6	9.8	9.9	7.5	11.0	8.1	4.6	4.0	4.1	1.7	2.9	23.7
7	7.3	7.8	6.8	6.8	7.8	5.3	4.4	1.0	2.9	2.9	29.2
8	6.0	4.3	7.7	3.4	7.7	5.1	4.3	4.3	2.6	6.0	35.9
Treated											
96-1	10.3	16.4	9.7	6.1	7.7	7.7	5.7	2.2	1.8	2.2	4.2
2	14.7	13.8	10.1	6.9	6.0	4.1	3.2	2.8	4.6	4.1	7.8
3	9.1	13.1	12.8	4.0	5.1	3.7	2.2	2.2	1.1	4.0	4.7
4	20.0	14.2	11.6	3.2	4.5	3.9	1.9	2.6	2.6	1.9	8.4
95-4	18.6	15.6	8.6	7.9	6.8	5.3	4.6	1.5	1.5	2.0	5.5
5	10.6	15.2	7.5	9.3	6.7	4.4	3.1	3.3	3.1	2.8	9.8
6	10.8	14.4	11.7	7.7	7.3	5.1	5.7	4.4	2.6	3.1	11.3
7	15.2	18.8	13.2	10.6	7.1	5.3	2.8	3.0	0.5	2.0	2.5
8	21.2	10.7	8.1	8.6	7.6	5.0	6.3	3.7	2.9	1.3	5.8
95-1*	20.1	13.9	9.0	7.8	5.4	2.5	1.8	2.7	1.5	0.1	2.2
2*	17.9	16.8	8.1	6.6	4.1	2.0	1.1	0.4	0.7	1.6	1.8
3*	15.0	14.5	10.1	7.9	3.7	5.1	3.9	2.1	1.7	0.5	2.6
93-1	17.4	15.1	8.8	7.1	5.7	4.0	4.3	4.0	1.4	1.7	3.7
2	10.8	15.6	13.2	6.6	8.1	5.1	3.3	1.5	1.2	2.4	7.5
3*	11.6	16.6	11.3	7.9	5.8	7.1	3.9	4.0	2.4	1.6	2.1
4	16.6	16.6	14.0	10.6	4.7	2.5	2.1	4.2	1.3	0.8	4.2
5	12.6	12.6	12.1	7.4	6.8	6.8	9.5	3.7	5.3	4.2	4.2

* Samples taken from 3-cm. depth; all others, surface samples.

There is, however, a considerable variation in figures obtained from surface samples, and this variation may be assumed to express soil heterogeneity,

which would be particularly marked in such small specimens as are used for thin sections. The variation was further increased by the treatment with the

TABLE 2a
Elten soil: pore-space percentages

THIN SECTION NUMBER	TOTAL LENGTH OF PORE SPACE AS A PERCENTAGE OF THE OVER-ALL LENGTHS EXAMINED										
	<160μ	160- 319μ	320- 479μ	480- 639μ	640- 799μ	800- 959μ	960- 1119μ	1120- 1279μ	1280- 1439μ	1440- 1599μ	>1600μ
Control											
94-1	8.2	2.1	1.3	0.4	0.3	0.1	0.2	...	0.2
2	6.2	2.4	0.9	0.5	0.2	0.1	0.1	...	0.1	...	0.1
3	6.0	1.5	0.8	0.6	0.3	0.1	0.1	...	0.2
4	3.8	1.2	0.5	0.1	0.1	0.1
5	3.1	0.5	0.6	0.2	0.1	0.1	0.1
90-1	13.2	3.8	1.3	0.5	0.6	0.2	0.1	...	0.2	0.1	0.2
2	7.3	2.1	1.3	1.0	0.5	0.5	0.3	0.2	...	0.1	0.1
3	13.2	4.6	0.5	0.5	0.8	1.6	0.5	0.3	0.3	...	0.5
4	10.0	3.1	2.0	0.4	1.3	0.6	0.1	0.2	0.1	0.1	0.3
5	11.0	5.6	1.8	1.3	0.6	1.6	0.3	0.6	0.2	0.3	1.2
6	6.8	2.5	1.5	0.2	0.5	0.2	0.1	0.1	0.1	0.2	0.4
7	7.2	3.6	1.5	1.4	0.6	0.7	0.8	0.2	0.2	0.2	1.2
8	11.8	4.0	2.8	0.5	1.0	0.2	0.5	...	0.5	0.3	1.3
Treated											
96-1	15.7	6.5	4.3	2.6	1.7	1.0	1.0	1.0	0.2	0.2	1.7
2	11.8	4.7	1.5	1.6	1.1	0.1	0.4	0.1	0.4
3	16.0	4.1	4.5	2.5	1.1	2.0	1.1	0.7	0.4	0.9	1.3
4	14.4	4.3	2.4	1.2	1.2	0.2	0.5	0.5	0.5
95-4	16.5	3.3	1.5	0.4	0.3	0.1
5	15.2	4.4	2.1	1.2	0.7	0.2	0.2	0.2	0.1
6	11.6	1.9	0.8	0.6	0.5	0.2	0.1	0.0	0.1	...	0.0
7	14.1	2.5	1.3	0.4	0.3	0.1	0.1	0.1
8	15.4	1.7	0.8	0.5	0.1	0.1	0.1	...	0.3
95-1*	20.1	6.2	3.4	1.3	0.7	0.6	0.3	...	0.2	0.2	...
2*	22.9	7.2	3.0	2.6	1.4	0.4	...	0.3	0.2	0.2	0.6
3*	19.2	8.0	2.6	1.2	0.8	0.7	0.4	0.2	0.4	0.1	0.1
93-1	15.0	4.5	2.7	1.9	1.2	0.8	0.1	0.2	0.1	...	0.3
2	11.8	4.6	4.3	0.8	1.3	0.7	0.5	0.2	0.1	...	0.2
3*	12.4	6.5	2.9	2.3	0.7	0.1	0.4	0.1	...	0.1	0.3
4	13.9	4.2	1.9	0.9	0.4	0.2	0.2	0.1	...	0.1	0.1
5	12.2	2.6	1.3	0.6	0.1	0.2	0.5	...	0.1

* Samples taken from 3-cm. depth; all others, surface samples.

humus material. It seems reasonable to suppose that this increase in variability is due to the impossibility of even distribution of the amendment on the field scale, although the humus material was finely divided.

A mean error of about 10 per cent resulted from the examination of 24 sections from soil treated with the humus product to afford a good structure; but only 15 sections of the untreated soil, which had a poor structure, needed to be examined to reduce the mean error to about 10 per cent.

TABLE 3

Etten soil: solid-soil, pore-space, and large crumb percentages and quotients

THIN SECTION NUMBER	SOIL	PORE SPACES	CRUMBS >1600 μ	QUOTIENT
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	
Control				
94-1	87.3	12.7	22.2	57.3
2	89.4	10.6	32.3	32.8
3	90.5	9.5	31.1	30.4
4	94.2	5.8	31.7	18.2
5	95.4	4.6	37.8	12.1
90-1	79.6	20.4	8.5	240.0
2	86.5	13.5	27.2	49.5
3	77.0	23.0	37.7	60.9
4	81.7	18.3	30.6	59.7
5	75.5	24.5	24.2	101.4
6	87.5	12.5	23.7	52.8
7	82.2	17.8	29.2	61.0
8	87.2	22.8	35.9	63.5
Treated				
96-1	64.1	35.9	4.2	860.9
2	78.3	21.7	7.8	276.9
3	65.3	34.7	4.7	731.9
4	74.9	25.1	8.4	298.7
95-4	77.8	22.2	5.5	404.0
5	75.7	24.3	9.8	248.0
6	84.2	15.8	11.3	140.7
7	81.2	18.8	2.5	741.3
8	81.1	18.9	5.8	328.0
95-1*	67.1	32.9	2.2	1470.0
2*	61.2	38.8	1.8	2144.0
3*	66.2	33.8	2.6	1289.0
93-1	73.2	26.8	3.7	725.4
2	75.5	24.5	7.5	325.1
3*	74.3	25.7	2.1	1219.0
4	77.8	22.2	4.2	521.4
5	82.1	17.9	4.2	424.2

* Samples taken from 3-cm. depth; all others, surface samples.

Table 4 and figure 2 give results for the Goes soil *in situ*. This is one of the stickiest soils in Holland. At Goes, samples were taken from the surface and at a depth of 25 cm. In the Goes soil, as in the Etten soil, application of the Van Reesema humic material brought about an improvement in structure.

Figure 3 shows the results from the Goes soil transported to Wageningen.

It will be seen that mere transport produced an improvement in structure. The improvement effected by the organic product, applied to the soils at Wageningen, is of the same order as that attained by its application to the soils *in situ* in the field.

TABLE 4
Summary of Goes soil data

	SOIL	PORE SPACES	CRUMBS >1600 μ	QUOTIENT
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	
Goes treated, surface.....	83.8	16.2	21.4	86.5 \pm 10.0
Goes control, surface.....	90.7	9.3	40.4	27.3 \pm 3.7
Goes control, at about 25-cm. depth.....	98.9	1.1	69.1	1.6 \pm 0.4

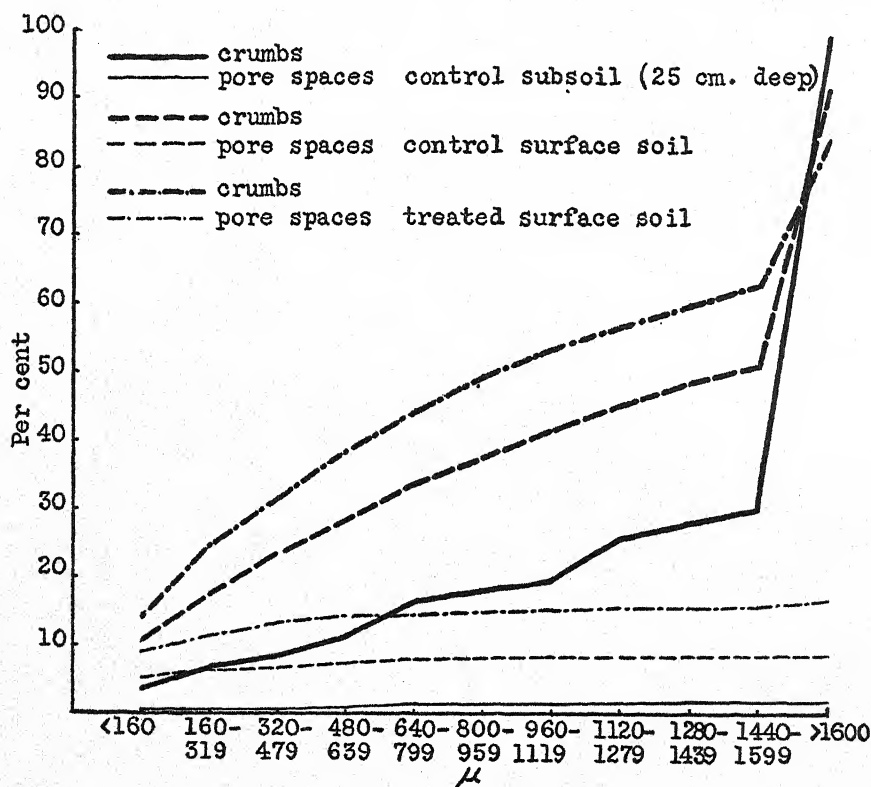


FIG. 2. GOES SOIL, SURFACE AND SUBSOIL, PERCENTAGES OF CRUMBS AND OF PORE SPACES

From the formula standard error of the mean = $\left(\frac{\sum v^2}{n(n-1)}\right)^{\frac{1}{2}}$ applied to the data in table 3, the average quotient of thin sections of untreated Etten soil was found to be 64.6 ± 15.9 , giving a mean error of 24.5 per cent. This

relatively large percentage is due to inclusion of only 13 sections for examination. For the treated Etten surface soil a mean quotient of 463.6 ± 63.6 was found (error, 13.7 per cent); the quotient in the treated soil at a depth of 3 cm. was 1530.5 ± 211 (error, 13.8 per cent). Here again, the relatively large errors were due to the examination of insufficient numbers of sections.

The significant difference in quotients representing good and bad structures can be evaluated from the square root of the sums of squares of the quotients.

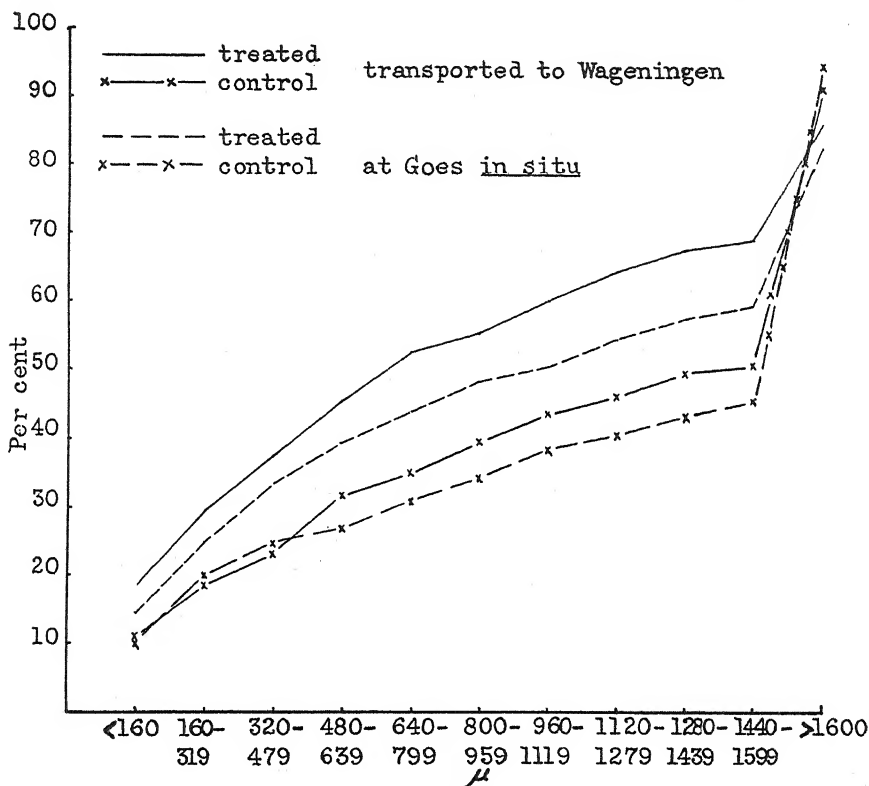


FIG. 3. GOES SOIL, IN SITU AND TRANSPORTED, PERCENTAGES OF CRUMBS

When this is applied to the results of the Etten surface soil, a value of 399 ± 65.6 is obtained, showing an improvement in structure of the order of 100 per cent, due to application of the humic material. A similar result was obtained on the Goes soil, and as already mentioned, on the soils taken to Wageningen and treated there.

The applicability of the thin-section method of examining soil structure and also the suitability, for numerical expression of the structure of a soil, of the methods described in this paper are still being investigated and reviewed in this laboratory.

SUMMARY

A method for visually observing the structure of a soil, with the aid of a microscope, and methods for numerically expressing pore space and/or the proportion of solid material—methods developed from Kubiena's thin-section technic—are described.

On the theory that soil structure depends on the size of pore spaces as well as on the size of the particles or crumbs of solid material, a ratio has been proposed which takes into account the pore space and the larger aggregates or crumbs. For this ratio the term *soil quotient* is proposed.

The quotient is believed to express the soil structure. Comparison of the quotients of soils appears to be capable of revealing differences in structure as small as those produced by the application on the field scale of a humic amendment.

PLATE 1

THIN SECTIONS OF THE ETTEN AND GOES SOILS

(Magnification about 5X)

FIG. 1. Surface soil from Etten in its untreated condition—"continental" structure

FIG. 2. The same surface soil as figure 1 after treatment with the "Van Reesema" humus material—"archipelago" structure

FIG. 3. The same soil as figure 2, but 3 cm. deeper and therefore less influenced by rainfall—typical good "archipelago" structure

FIG. 4. The Goes surface soil, in its untreated condition—"continental" structure

FIG. 5. The Goes subsoil from 25 cm. depth in its untreated condition—"heavy continental" structure

FIG. 6. The same soil as figure 4 after treatment in the field with the "Van Reesema" humus material—"archipelago" structure

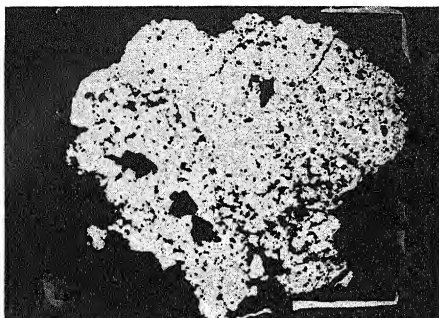


FIG. 1

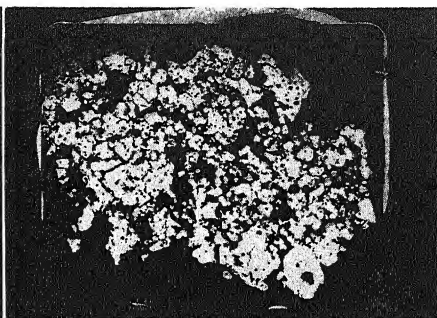


FIG. 2

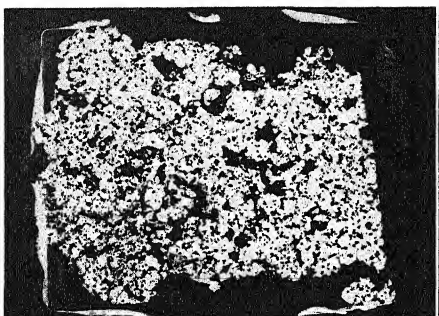


FIG. 3

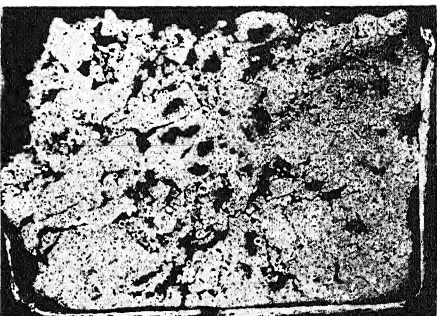


FIG. 4

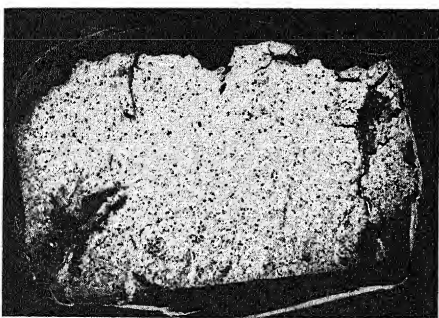


FIG. 5

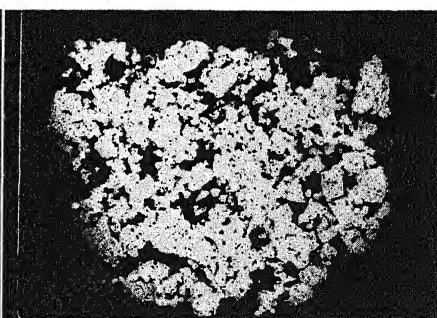


FIG. 6



BACTERIOPHAGE OF RHIZOBIA IN RELATION TO SYMBIOTIC NITROGEN FIXATION BY ALFALFA¹

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The lytic power of *Rhizobium*-phage isolated from root nodules of various legumes has been demonstrated in laboratory studies by a number of investigators (7, 9, 11, 12, 13). The fact that this lytic principle is present in and may be isolated from certain "alfalfa sick" or "fatigued" soils is indicated convincingly by the studies of Demolon and Dunez (1, 2, 3) and by previous work (13) in our laboratories. Demolon and Dunez concluded that the diffusion of *Rhizobium*-phage in alfalfa soils may cause the disappearance of the homologous rhizobia in the root zone and a cessation of bacterial symbiosis, with the result that alfalfa is forced to grow like a nonleguminous plant. They observed that the exterior symptoms of "alfalfa sickness" or "fatigue" in alfalfa fields are always associated with the appearance of phage in the plants and that the degree of "alfalfa sickness" is closely correlated with the amount of lytic principle in the nodule and plant tissue. Previous studies in our laboratory (13) indicated that the presence of *Rhizobium*-phage in various irrigated central Washington soils occupied by 3- to 5-year-old stands of alfalfa coincided with decreased alfalfa yields, and that the activity of this lytic principle might be responsible for a reduction in symbiotic nitrogen fixation and consequently a reduction in yields of alfalfa. The results of further studies of the behavior of alfalfa in relation to *Rhizobium*-phage are reported in this paper.

EXPERIMENTAL PLAN AND PROCEDURE

In the spring of 1936 a suitable quantity of soil from the surface 10-inch layer and a number of 4-year-old alfalfa plants were obtained from an irrigated alfalfa field which in the previous year was found to be infected by a phage causing lysis of *Rhizobium meliloti*. This is soil 3 in a previous publication (13). The soil was thoroughly mixed before its moisture content had reached the wilting percentage. It was then passed through a 4-mesh screen, to remove

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the coarse roots and other coarse organic substances, and placed in 12-quart pails the inside surfaces of which were protected by two coats of Gilacoat asphaltite. After receiving the following treatments in triplicate pails, alfalfa was seeded, and in addition, three of the 4-year-old alfalfa plants obtained from the field were planted in each of the pails 11 to 15 inclusive:

Pail No.	Treatments
1, 2, 11	No treatment
3, 4, 12	20 cc. suspension of <i>Rh. meliloti</i> , strain A-2 (susceptible)
5, 6, 13	20 cc. suspension of <i>Rh. meliloti</i> , strain A-2, and 30 cc. of an A-2 homologous phage
7, 8, 14	20 cc. suspension of <i>Rh. meliloti</i> , strain A-1 (resistant)
9, 10, 15	20 cc. suspension of <i>Rh. meliloti</i> , strain A-1, and 30 cc. of an A-2 homologous phage

The primary purpose of this experiment was to study the effect of the *Rhizobium*-phage in the soil on the growth of alfalfa, with and without the addition of *Rhizobium*-phage filtrates, and of susceptible and resistant strains of *Rh. meliloti*.

The lytic agent used for inoculation was that isolated from soil 3 in the previous study (13). The *Rh. meliloti* strains A-1 and A-2 were the ones which proved to be resistant and susceptible, respectively, to the action of *Rhizobium*-phage in fluid cultures and on agar plates in the same study. The introduction of old alfalfa plants in pails 11 to 15 inclusive was intended mainly to assure perpetuation of the phage in case it might be unable to survive for more than a few weeks in the absence of homologous nodules or host plants.

The pails were kept in the greenhouse from the time of seeding in May 1936 until July 1937, and the alfalfa was cut when the plants were at the half-bloom stage of maturity. No attempt was made to produce a crop during the short winter days when a low temperature was maintained in the greenhouse to curtail top growth. Following the first cutting of alfalfa in 1937, the pails were moved out of doors, and all succeeding growth occurred under natural climatic conditions. No yield data were recorded in 1936 and 1937.

The presence of phage in the soil was determined by the following method: Representative samples were obtained aseptically in August 1937 and in November 1938 by means of borings taken with a $\frac{1}{2}$ -inch soil auger. Four borings from each pail were composited at each sampling. The composite samples were first passed through a 16-mesh sieve, and then 100-gm. portions of the sifted soil were placed in flasks containing 200 cc. of fluid medium [medium 4 of Laird (11)], incubated at 28°C. for 48 hours, filtered through paper, and the resulting filtrates passed through Berkefeld N candles. Two-cubic-centimeter portions of the filtrates were transferred to test tubes containing 10-cc. quantities of fluid 24-hour cultures of *Rh. meliloti* (strain A-2) grown in medium 5 of Laird (11). These cultures were incubated at 28°C. for 48 hours, when observations were made for lysis. They were then filtered through sterilized Berkefeld N candles, and 2-cc. portions of the filtrates were added to 24-hour

fluid cultures of *Rh. meliloti*, whereupon the process of incubation, observation for clearing, and filtration was repeated. Serial transferring in this manner was repeated until complete clearing of the fluid cultures was observed within the 48-hour incubation period.

The presence of *Rhizobium*-phage in the root nodules was determined by the following procedures: First, representative nodules obtained from the roots of healthy and phage-infected alfalfa plants were washed and crushed. As much as possible of the nodule tissue being discarded, slide smears of the crushed material were prepared and stained for 10 minutes with a 1:1000 solution of gentian violet in accordance with the procedure of Demolon and Dunez (1). Second, representative nodules obtained from the roots of healthy and phage-infected alfalfa plants were washed, sterilized in a 1:500 HgCl_2 solution for 15 minutes, then thoroughly rinsed in sterilized water, and finally cut to pieces aseptically. Several pieces of the nodules from both the healthy and the phage-infected plants were placed aseptically in agar plates on 24-hour streaks of *Rh. meliloti*, (strain A-2) to determine the lytic properties of the nodule material.

Following the first cutting of the alfalfa on June 14, 1938, pails 2, 4, 6, 8, and 10-15 received ammonium sulfate (21 per cent N) at the rate of 600 pounds per acre; and pails 11-15 received, in addition, treble superphosphate (45 per cent P_2O_5) 200 pounds per acre and potassium sulfate (50 per cent K_2O) 150 pounds per acre. Following the second cutting of alfalfa on July 28, 1938, ammonium nitrate (35 per cent N) supplying a quantity of nitrogen equivalent to 400 pounds of ammonium sulfate per acre was applied to pails 2, 4, 6, 8, and 10-15. The purpose of these treatments was to determine their effect on the growth of the alfalfa, which had declined in vigor in 1937 and especially in the spring of 1938.

Following the third cutting of alfalfa on September 13, 1938, representative plants were carefully removed from the various pails for a thorough examination for bacterial wilt and other related alfalfa diseases.

EXPERIMENTAL RESULTS

Effect of Rhizobium-phage on growth of alfalfa

The young alfalfa produced in the greenhouse in 1936 was normal in growth and vigor. No yield data were recorded, as no effect from inoculation or *Rhizobium*-phage treatments was discernible. The first crop in 1937, also produced in the greenhouse, was damaged considerably by red spider, but the following two cuttings, which were produced out of doors, were free from insect pests. The growth of the first of these two cuttings appeared to be normal, whereas that of the second cutting lacked vigor and failed to attain the proper height. In neither case were differences in growth and vigor discernible as a result of inoculation or *Rhizobium*-phage treatment, and therefore, no yield data were recorded.

The presence of *Rhizobium*-phage in the soil was readily demonstrated in the samples taken from all the pails in August following the first cutting grown out of doors. Without exception, the 24-hour fluid cultures of *Rh. meliloti* (strain A-2) were lysed completely in 24 hours after the first serial transfers of the soil filtrates were made. The potency of the lytic principle was of the same high order of magnitude in all except the untreated soil, in which it was slightly slower in action.

TABLE 1
Yields of alfalfa in Rhizobium-phage infected soils in 12-quart pails
Air-dried weight in grams

PAIL NUMBER	TREATMENT	FIRST CUTTING 6/14/38	SECOND CUTTING 7/23/38	THIRD CUTTING 8/13/38	AVERAGE OF SECOND AND THIRD CUTTINGS	FIRST CUTTING 1939
1	None	18.0	19.5	14.5	17.0	11.2
3		15.0	13.5	8.5	11.0	13.2
5		18.5	15.0	12.0	13.5	13.0
7		24.5	17.0	13.0	15.0	16.4
9		26.5	19.5	13.5	16.5	13.9
Ave.		20.5	16.9	12.3	14.6	13.5
2	N*	20.5	35.5	42.5	39.0	19.0
4		19.0	40.0	42.0	41.0	15.0
6		18.0	40.5	37.5	39.0	16.2
8		26.0	45.5	50.5	48.0	21.2
10		21.0	39.0	38.5	38.7	23.0
Ave.		20.9	40.1	42.2	41.1	18.9
11	NPK†	31.5	46.5	51.6	49.0	29.4
12		34.5	54.5	39.5	47.0
13		16.5	40.0	47.0	43.5	26.9
14		37.5	50.0	53.8	51.9	32.4
15		17.0	42.5	40.0	41.3	28.7
Ave.		27.4	46.7	46.4	46.5	29.3

* N applied at the rate of 126 pounds per acre for the second cutting and at the rate of 84 pounds per acre for the third cutting in 1938 only.

† N applied at the same rates as in the N treatment, P applied at the rate of 90 pounds P_2O_5 , and K at the rate of 75 pounds of K_2O per acre for the second cutting in 1938 only.

The growth of the first cutting of alfalfa in 1938 was characterized by lack of vigor, light green color, and less than normal height, the symptoms being similar to those observed by Demolon and Dunez (1) in "fatigued" alfalfa fields and by Vandecaveye and Katznelson (13) in certain 3- and 4-year-old stands of alfalfa on *Rhizobium*-phage infected irrigated soils in central Washington. The thought that the phage, which proved to be abundantly active in the soil, might be interfering with normal symbiosis and consequently

causing a reduction of the supply of available nitrogen for the alfalfa, or that one or more of the other principal essential plant nutrients might be deficient, suggested further investigation. The basic plan followed was to make applications of various fertilizers immediately following the first cutting in June and the second cutting in July, as outlined in the experimental procedure. The substitution of ammonium nitrate for ammonium sulfate following the second cutting was to prevent the accumulation of excessive concentrations of residual sulfate ions. The phosphate and potash fertilizer treatments were not repeated after the second cutting.

The addition of nitrogen to the soil resulted in greatly improved color, vigor, and top growth of the alfalfa, as may be noted in plate 1. The magnitude of the response to nitrogen is well illustrated by the yield data in table 1. The application of nitrogenous fertilizer alone caused more than a twofold increase in average yield for the second cutting and more than a threefold increase in average yield for the third cutting. The averages of the two cuttings combined indicate that the addition of nitrogen resulted in approximately a threefold increase in yield. Additional stimulation in growth appeared to result from the application of phosphate and potash fertilizers, but this effect was not pronounced, since the soils in this series of pails, which bore the old alfalfa plants together with the new seeding, produced slightly better average yields previous to the fertilizer applications than did the other two series of pails. The beneficial effect of the phosphate and potash fertilizers seemed to be more distinct in 1939, however, as may be noted from the yields of the first cutting of alfalfa, which was produced without further additions of fertilizers.

As in 1936 and 1937, no significant differences in the appearance or yield of the alfalfa resulted from the *Rhizobium*-phage treatments or from the inoculation with either the phage-susceptible strain, A-2, or the phage-resistant strain, A-1, of *Rh. meliloti*, both of which were capable of producing an abundance of nodules and of benefiting the growth of alfalfa in separate sterilized sand and soil culture tests. *Rhizobium meliloti* strain A-1, which proved to be resistant to lysis in fluid cultures (13), failed to stimulate the growth of alfalfa when it was used as an inoculant in this *Rhizobium*-phage infected soil. The principal, if not the only, limiting factor in the growth and yield of alfalfa appeared to be available nitrogen. When sufficient nitrogenous fertilizer material was added to the soil, large yields were obtained. When the nitrogen fertilizer was no longer added, as in the case of the first crop in 1939, the yields dropped to about the same level as that which prevailed before the fertilizers were applied.

Distribution and activity of Rhizobium-phage

The soils in the pails were sampled in November 1938. A few weeks later, samples of representative nodules, which were found to be fairly abundant on the roots of the alfalfa plants, were obtained from the various pails and also from the roots of alfalfa plants produced on soils that were not infected by

Rhizobium-phage. The soil and root-nodule samples were treated and analyzed in accordance with the methods described in the experimental procedure. The purpose of this analysis was to study the distribution of the phage and the nature of its activity in relation to the symbiosis normally occurring between the *Rh. meliloti* in the nodules and its homologous host plant.

Positive tests for *Rhizobium*-phage were readily obtained from all the soil samples. In all cases the first serial transfer of the soil filtrate caused complete lysis of 24-hour fluid cultures of *Rh. meliloti*, strain A-2, within 24 hours of incubation at a temperature of 28°C. The potency of the phage was essentially the same as it was when the soils were sampled in 1937, but distinctly greater than was demonstrated by the soil samples taken from the field in 1935 (13), when a second serial transfer was required to effect complete lysis in 24 hours. Lysis was uniform with respect to degree and rate, except that the time factor was slightly increased for the lysis resulting from the filtrate of the soils which had not received *Rhizobium*-phage or *Rh. meliloti* by inoculation. This slight difference probably is not significant. The remarkable uniformity in vigor and potency of the phage regardless of the previous treatments of the soil in the various pails is an indication that the addition of the isolated *Rhizobium*-phage or of the susceptible or resistant *Rh. meliloti* to the soil exerted no appreciable influence on the activity of the phage in the soil. This is further substantiated by the crop yield data in table 1, which fail to show any significant effect as a result of these treatments.

The action of phage on the bacteria in the nodules was effectively demonstrated by stained slide smears prepared from crushed nodule material. Demolon and Dunez (4) used this method successfully in a study of the appearance of *Rhizobium*-phage and the progress of its activity in alfalfa fields. A typical example of the slide smears is shown in plate 2, figure 1. The rhizobia in the phage-infected nodules, which may appear to be sound and healthy, take the stain poorly and are characterized by a high degree of cell disintegration and granulation. The rhizobia in noninfected, active, sound nodules fix the stain energetically, and relatively few of the cells are disintegrated or granular in form. Evidently bacteriolysis took place in the *Rhizobium*-phage infected nodules in this experiment. It seems quite probable that although a fairly large number of nodules may be produced in the presence of a potent *Rhizobium*-phage, as was actually the case in this experiment, lysis may be so energetic as to impede symbiotic nitrogen fixation to the extent of reducing the yield to less than one third that of noninfected plants in normal condition. That similar decreases in yields may result from phage infection of alfalfa under field conditions is indicated by the work of Demolon and Dunez (4, 5).

The presence of phage in the nodules and its destructive action on *Rh. meliloti* were further demonstrated by placing pieces of infected and noninfected nodules on 24-hour streaks of *Rh. meliloti*, strain A-2, on agar plates. The results are illustrated in plate 2, figure 2. Nodules obtained from pails 2, 4,

6, 8, and 10 all indicated the presence of *Rhizobium*-phage by distinct lysis in the area bordering the nodule tissue. This is indicated in streak 2 (pl. 2, fig. 2). No lysis occurred on streak 1, on which pieces of the healthy, non-infected nodules were placed; streak 3, which received one loopful of homologous bacteriophage isolated from soil 3, was lysed almost completely.

Although the presence of phage in the root nodules and its destructive action on the nodule bacteria were clearly demonstrated, its effect in reducing or checking symbiotic nitrogen fixation, though shown indirectly by greatly increased growth of the host plant in response to applications of nitrogen fertilizers, could have been complicated by other factors. The plant symptoms caused by *Rhizobium*-phage, bacterial wilt, and virus, which may coexist at times, are easily confused. The symptoms of bacterial wilt, alfalfa dwarf, and witches' broom, as described by Jones and Weimer (10), Weimer (14), and Edwards (6), are characterized by certain aspects which are not unlike those exhibited by advanced stages of *Rhizobium*-phage. Bacterial wilt and witches' broom have been identified in numerous 3- and 4-year-old stands of alfalfa on irrigated soils in central Washington. The destructive effect of bacterial wilt, like that of bacteriophage, becomes especially pronounced in the third and fourth year after the alfalfa is seeded, and bacterial wilt has been thought to be one of the principal causes of the declining yields of alfalfa in those fields. To make sure that the *Rhizobium*-phage problem in our experiment was not complicated by other disease factors, a number of plants, including both tops and roots, were carefully removed from the various pails and thoroughly examined. No symptoms of bacterial wilt, alfalfa dwarf, or witches' broom were discovered. Thus the only factor which can logically account for the poor growth of alfalfa in this experiment is *Rhizobium*-phage.

DISCUSSION

Although the results of our experiments indicate definitely that the lytic action of *Rhizobium*-phage on 3-year-old alfalfa stands may result in a reduction of 60 to 70 per cent of the yields normally obtained from noninfected healthy stands, it would be premature, on the basis of the foregoing data, to suggest the full significance of this lytic principle in relation to symbiotic nitrogen fixation.

Rhizobium-phage which various investigators, including Gerretsen et al. (7) and Hitchner (8), once believed to be specific, was found by Laird (11, 12) and by Demolon and Dunez (4) to be nonspecific, although the last-named workers observed that phage isolated from alfalfa fields was the most potent and that the various phages studied were generally more potent for rhizobia species of homologous host plants than for rhizobia species of heterogeneous leguminous plants. The nonspecific property of these phages suggests a tendency toward continuous propagation in infected soils bearing different species of legumes in succession and also continued interference with symbiotic nitrogen fixation.

Another complicating factor in the *Rhizobium*-phage problem is the unequal susceptibility of different strains of the same species of *Rhizobium*. Laird (12) observed marked variation in susceptibility among different strains isolated from the same nodule and also a change in susceptibility of certain strains. Demolon and Dunez (5) have succeeded in developing a lyso-resistant strain of *Rh. meliloti* which appears to be capable of maintaining normal symbiosis for several years when alfalfa seed properly inoculated with this organism is planted in "fatigued" soil. The possibility that rhizobia may develop various degrees of lyso resistance under natural conditions in the soil suggests itself. Moreover, nodules developing on different varieties of the same species of legumes may not be equally susceptible to phage infection. Demolon and Dunez (2) observing 14 varieties of alfalfa growing side by side on "fatigued" soil found marked varietal differences in susceptibility to the effects of this lytic principle. Reliable evidence now available from the results of field experiments in various parts of this country indicates that certain varieties of alfalfa are highly resistant to bacterial wilt. In a field plot experiment on irrigated soil in central Washington, 4- and 5-year stands of certain of these bacterial-wilt resistant varieties are still producing normal high yields, whereas bacterial-wilt susceptible varieties growing alongside the resistant ones began to lose their vigor and produced rapidly declining yields the third year after the alfalfa was seeded. The soil in both cases was analyzed in 1938 and was found to be infected by a potent *Rhizobium*-phage. Is it possible that certain bacterial-wilt resistant varieties of alfalfa may be phage resistant also? This and the other factors mentioned require much additional study and need to be correlated properly before the full significance of *Rhizobium*-phage and its relation to symbiotic nitrogen fixation can be ascertained. A controlled pot experiment now in progress, including the use of phage-susceptible and phage-resistant strains of *Rh. meliloti* as well as bacterial-wilt resistant and susceptible varieties of alfalfa, was designed further to elucidate some of these factors.

SUMMARY

Rhizobium-phage infected soil placed in 12-quart pails and treated variously with *Rhizobium*-phage in fluid cultures and with phage-resistant and phage-susceptible strains of *Rh. meliloti* was used for the production of alfalfa during $3\frac{1}{2}$ years.

The growth and yields of alfalfa were normal in 1936, the first year, but began to decline toward the end of 1937.

The addition of nitrogenous fertilizer in 1938 to two series of the triplicate pails following the first cutting of alfalfa, which lacked vigor and yielded poorly regardless of previous treatments, resulted in greatly improved vigor and nearly a threefold increase in yield. A second application of nitrogenous fertilizer following the second cutting of alfalfa had even more striking beneficial effects. Phosphate and potash fertilizers added to one of the fertilized series as supplements to nitrogen had no significant beneficial effects.

Samples of soil obtained from the various pails in 1937 and in 1938 readily revealed the presence of a very active phage, the potency of which was uniformly high regardless of previous applications of *Rhizobium*-phage or phage-resistant and phage-susceptible *Rh. meliloti* to the soil. When the phage-resistant strain of *Rh. meliloti* was used as a soil inoculant, it had no appreciable beneficial influence on the growth of the alfalfa.

The presence of phage in the nodules of plants grown on infected soil was demonstrated by lysis of those parts of 24-hour agar streak cultures of *Rh. meliloti* in contact with such nodule tissue and by disintegration and granulation of rhizobia cells as revealed by stained slide smears of crushed nodule material.

A careful examination of representative alfalfa plants taken from the variously treated soils in the pails failed to disclose symptoms of bacterial wilt, alfalfa dwarf, or witches' broom. These factors eliminated, the only logical factor accountable for the poor growth of alfalfa in this experiment is the action of *Rhizobium*-phage resulting in lysis of *Rh. meliloti* and in a consequent drastic reduction of symbiotic nitrogen fixation.

Some of the complicating factors that should be considered in connection with the *Rhizobium*-phage problem are discussed briefly.

REFERENCES

- (1) DEMOLON, A., AND DUNEZ, A. 1935 Recherche sur le rôle du bactériophage dans la fatigue des luzernières. *Ann. Agron.* 5: 89-111.
- (2) DEMOLON, A., AND DUNEZ, A. 1936 La fatigue des sols. Cause et remèdes. *Compt. Rend. Acad. Sci. [Paris]* 202: 1704-1706.
- (3) DEMOLON, A., AND DUNEZ, A. 1936 Nouvelles observations sur le bactériophage et la fatigue des sols cultivés en luzerne. *Ann. Agron.* 6: 434-455.
- (4) DEMOLON, A., AND DUNEZ, A. 1938 Symbiose bactérienne et cultures légumineuses. *Ann. Agron.* 8: 220-237.
- (5) DEMOLON, A., AND DUNEZ, A. 1939 Sur la lyso-résistance du *B. radicola* et son importance pratique. *Trans. 3rd Comn. Internatl. Soc. Soil Sci.* A: 39-42.
- (6) EDWARDS, E. T. 1936 Witches' broom disease of lucerne. N. S. Wales Dept. Agr. Sci. Bul. 52.
- (7) GERRETSEN, F. C., et al. 1923 Das Vorkommen eines Bakteriophagen in den Wurzelknöllchen der Leguminosen. *Centbl. Bakt.* (II) 60: 311-316.
- (8) HITCHNER, E. R. 1930 The isolation of a bacterio-lytic principle from the root nodules of red clover. *Jour. Bact.* 19: 191-201.
- (9) ISRAILSKY, W. P. 1926 Phänomen d'Herrelle und Pflanzenkrebs. *Ber. Bakt. Agron. Sta. Moskow.* 24: 143-157.
- (10) JONES, F. T., AND WEIMER, J. L. 1928 Bacterial wilt and winter injury of alfalfa. U. S. Dept. Agr. Cir. 39.
- (11) LAIRD, D. G. 1932 Bacteriophage and the root nodule bacteria. *Arch. Microbiol.* 3: 159-193.
- (12) LAIRD, D. G. 1933 A study of strains of rhizobia with particular reference to the bacteriophage. *Proc. World's Grain Exhib. and Conf.* (Canada) 2: 362-369.
- (13) VANDECAVEYE, S. C., AND KATZNELSON, H. 1936 Bacteriophage as related to the root nodule bacteria of alfalfa. *Jour. Bact.* 31: 465-77.
- (14) WEIMER, J. L. 1931 Alfalfa dwarf, a hitherto unreported disease. *Phytopath.* 21: 71-75.

PLATE 1

SECOND CUTTING (HALF-BLOOM STAGE) OF A 3-YEAR-OLD STAND OF ALFALFA ON
Rhizobium-PHAGE INFECTED SOIL

FIG. 1. No fertilizer treatment

FIG. 2. Ammonium sulfate at the rate of 126 pounds of N per acre

FIG. 3. Ammonium sulfate, treble superphosphate, and sulfate of potash at the rates of 160 pounds of N, 90 pounds of P_2O_5 , and 75 pounds of K_2O per acre respectively

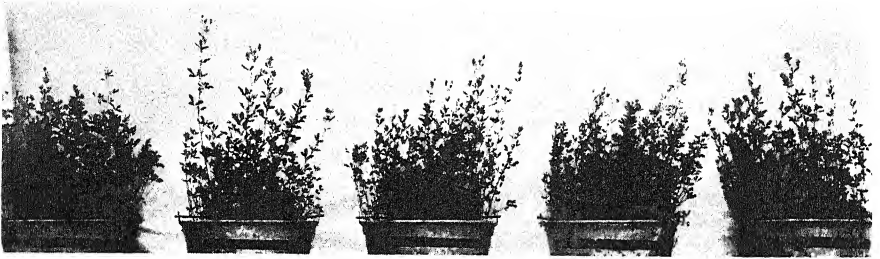


FIG. 1

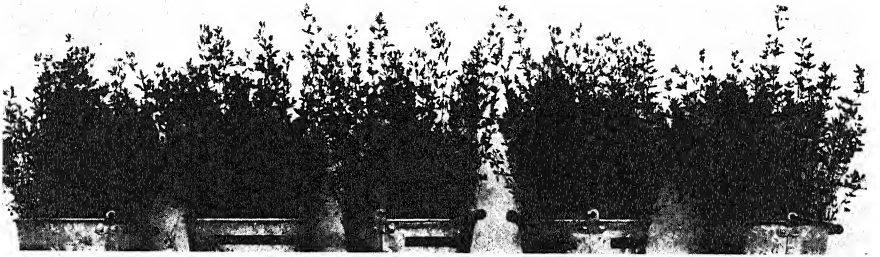


FIG. 2



FIG. 3

PLATE 2

FIG. 1. Stained preparations on *Rh. meliloti* cells in crushed nodules. Left—*Rh. meliloti* from noninfected, healthy nodules (approx. $\times 1600$); Right—*Rh. meliloti* from *Rhizobium*-phage infected nodules (approx. $\times 1600$)

FIG. 2. Lysis of *Rh. meliloti*, strain A-2, by *Rhizobium*-phage

1. Streak of *Rh. meliloti* with pieces of noninfected, healthy nodules
2. Streak of *Rh. meliloti* with pieces of *Rhizobium*-phage infected nodules
3. Streak of *Rh. melliloti* streaked with one loopful of *Rhizobium*-phage isolated from soil 3 (13)
4. Untreated streak of *Rh. meliloti*

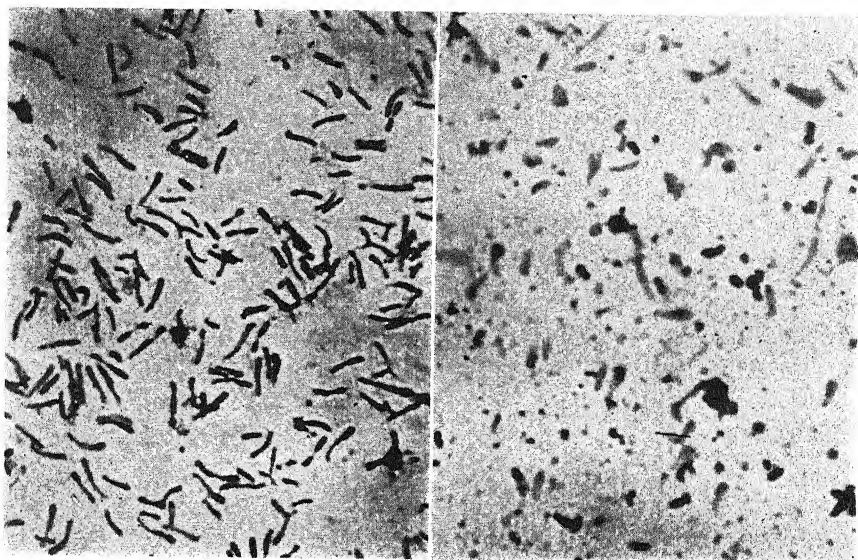


FIG. 1

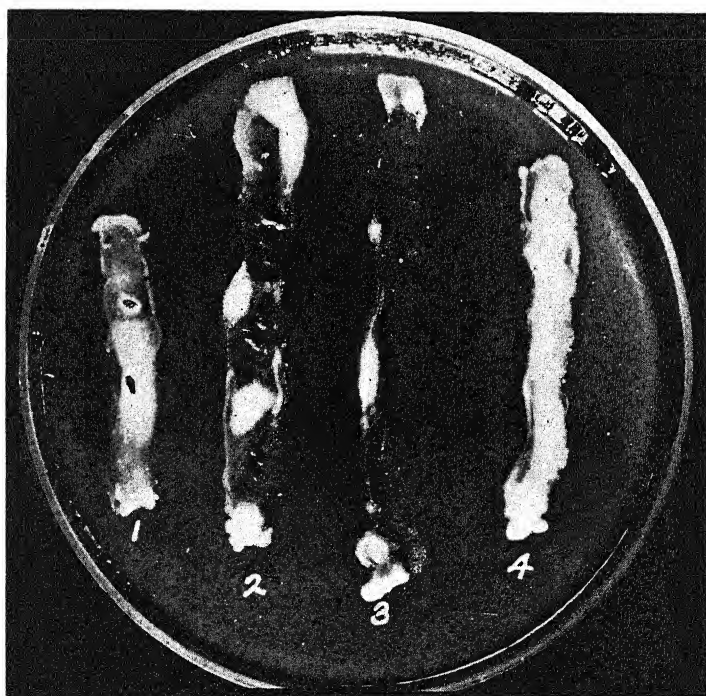


FIG. 2

INFLUENCE OF MICROORGANISMS ON SOIL AGGREGATION AND EROSION¹

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Considerable attention has been focused recently upon the problem of soil conservation, as a result of the efforts of both the Federal and State governments to control soil erosion and to reduce soil and water losses. The numerous investigations undertaken in different parts of the country have resulted in the accumulation of a large body of information on soil structure, as well as on the farm practices which lead to the conservation of the structure, on the one hand, and its destruction, on the other. The physical and chemical properties of the soil which determine its susceptibility to erosion have received particular consideration. Comparatively little attention has been paid, however, to the influence on soil conservation of the numerous microorganisms inhabiting the soil, whether exerted indirectly through the products of microbial activities or directly through the mass of cell substance synthesized.

Among the treatments which result in conserving soil from erosion, the introduction of organic matter has been found to play a particularly significant part. It has been shown (17) to reduce both soil and water losses and to decrease erosion by improving the physical properties of the soil. Furthermore, as a result of decomposition of the fresh organic matter added to the soil, humus is formed; this combines physically and chemically with the inorganic soil particles, forming more or less stable aggregates (1, 4, 7, 10, 12, 18, 23, 26). Bayer (2, 3, 19) reported a significant and positive correlation between the organic matter content and the degree of aggregation of soils belonging to the nonlateritic types. An increase in the number of water-stable aggregates increases the infiltration and percolation rates of the soil, and this in turn leads to a decrease in the runoff and soil losses (5, 11, 16, 20). The effect of organic matter on soil erosion was also measured by the change in percolation rate, infiltration rate, and aggregation (5, 20, 25). Organic matter was shown to reduce erosion by increasing infiltration and percolation due to the mechanical separation of the soil particles, by acting as tiny dams which retard the velocity of the runoff water, and possibly by increasing the water-absorbing and moisture-holding capacities of the soil. When used as a surface

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mulch, organic matter also breaks up the impact of the rain and prevents puddling of the soil, thus assisting the process of infiltration.

The incorporation of organic matter into the soil appears to give the greatest protection against erosion during the warm summer months soon after addition (17). This, it should be emphasized, is the period of the most rapid decomposition of organic matter by microorganisms. Various fungi develop rapidly and form a network of mycelium around the soil particles; this tends to bind the particles together (28, 29, 30). Bacteria, as well, develop and produce sticky substances, which tend to cement the soil particles together (8, 21, 22, 30). Certain products of decomposition of the plant and animal residues, as well as the cell substance synthesized by the microorganisms, exert various physical and chemical effects upon the inorganic soil colloids, resulting in the formation of more or less stable aggregates. Thus organic matter can be shown to have two distinct effects in soil conservation: When material like alfalfa tissue is incorporated into the soil it is immediately attacked by the soil population, with the formation of large quantities of microbial cell substance, which has one type of effect. On the other hand, when peat is added to the soil, its effect will be that of a resistant type of humus (29).

Because of differences in the nature and physiology of different organisms, the effects of the different members of the soil microbiological population upon the physical properties of the soil vary considerably. Since the plant materials added to the soil differ in chemical nature and result in the development of different groups of microorganisms, their effect upon soil conservation will not always be the same. Stephenson reported (27) that different types of organic material affect the physical properties of the soil differently. Metzger and Hide (14) found that alfalfa and sweet clover leave the soil in a state of greater aggregation than do several other crops.

Geltzer (8) came to the conclusion that during the process of mineralization of organic matter in soil, there is an accumulation of synthetic microbial substances which bring about the formation of soil aggregates. According to Kanivetz and Korneeva (9), certain organisms like *Trichoderma lignorum* added to decomposing straw bring about a marked increase in the concentration of water-resisting soil aggregates. These investigators spoke of the bacteria and fungi, in relation to soil structure, as "biochemical structure formers" and as "biochemical cement." It was previously reported from this laboratory (30) that fungi bring about a marked binding action by means of their mycelium, and that bacteria, through the production of slimy substances, also cause considerable binding of the finer soil particles. Peele (21) also concluded recently that the effect of microorganisms on soil structure appears to be much more important than has previously been considered.

In attempting to elucidate the role of microorganisms in soil conservation, the following factors were investigated: the functions of microbes as direct binding agents of soil particles; the production by microbes of various substances which aid in binding the soil constituents; the specific stage of decom-

position of organic matter at which it is most active in bringing about the binding or the aggregation of the soil particles; and the effects of additions of different types of organic matter to the soil.

In order to study the specific binding and aggregating effects of pure and of mixed cultures of microorganisms upon the soil, it was thought desirable in the first experiment to begin with a simple medium comparable to soil. For this purpose, sand-bentonite and sand-clay mixtures, which had previously (13) been found to be good artificial soil media for the growth of microorganisms, were employed.

EXPERIMENTAL

Methods

The binding and aggregating effects of mixed and pure cultures of microorganisms on soil and soil-like media variously treated in flasks were determined by the pipette and slope methods.

Pipette method. Fifty-gram portions of the material from the flasks were placed in water and allowed to stand for 15 minutes. The suspension was then transferred to a 250-cc. graduate, filled to the mark with water at 21°C., and shaken 10 times. The graduate was allowed to stand 1 minute, and 50 cc. of the liquid containing the particles remaining in suspension was pipetted out from a depth of 5 inches below the surface of the liquid. The aliquot was evaporated to dryness in weighed evaporating dishes, ignited, and weighed. The amount of material from the original 50-gm. portion remaining in suspension, after 1 minute, was calculated and is referred to as unbound material. The final calculation was made on the basis of 50 gm. of dry material. This method is used as a measure of the relative change in aggregation of the silt and clay constituents of the soil.

Slope method. Fifty-gram portions of the material were uniformly spread in the center of a small chute (14.5 by 4.5 inches) placed at a 5.5 per cent slope. The material was moistened with 50 cc. of water from a pipette, and 450 cc. of water siphoned from a liter flask was allowed to run over the mixture in approximately 2 minutes. Then 200-cc. aliquots containing the washed out particles were evaporated to dryness in weighed evaporating dishes, ignited, and weighed. The weight of the small particles washed from 50-gm. portions of the original mixture was taken as unbound material, and final calculation was made on 50 gm. of dry material.

Sand-bentonite mixture

The sand-bentonite mixture was prepared with 2.5 per cent bentonite and 97.5 per cent white river sand which had been screened through a 1-mm. sieve. The moisture-holding capacity of this mixture was 40 per cent. Sucrose and cellulose were added as sources of organic matter. The mixture, in 250-gm. portions, was placed in 500-cc. Erlenmeyer flasks, and the moisture content was brought to 55 per cent saturation by the addition of salt solution and, in

the sucrose series, of sugar solution. The flasks and their contents were sterilized at 15 pounds' pressure for 2 hours, inoculated in quadruplicate, and incubated at 28°C.

In the sucrose series, 40 cc. of the following salt solution was added to each flask:

(NH ₄) ₂ HPO ₄	8.32 gm.
CaCl ₂	0.82 gm.
MgSO ₄	0.82 gm.
K ₂ HPO ₄	6.25 gm.
FeSO ₄	Trace
H ₂ O to make.....	1000 cc.

The sucrose solution was added aseptically to give 2 per cent sugar on the basis of dry material, allowance being made for the water added. The flasks were inoculated as follows:

1. Control
2. *Rhizopus nigricans*
3. *Aspergillus niger*
4. *Azotobacter indicum*
5. *Pseudomonas fluorescens*
6. Soil infusion (1 cc. of a 1:5 soil suspension).

In the *Azotobacter*-inoculated flasks, the nitrogen was eliminated from the salt solution.

In the cellulose series, ground filter paper was added to make 2 per cent of the total dry material. The following salt solution (80 cc. per flask) was used:

NH ₄ NO ₃	11.25 gm.
K ₂ HPO ₄	3.33 gm.
MgSO ₄	2.00 gm.
NaCl.....	1.00 gm.
H ₂ O to make.....	1000 cc.

These flasks were inoculated as follows:

1. Control
2. *Trichoderma kőningi*
3. *Aspergillus fumigatus* + *T. kőningi*
4. *Penicillium* sp. + *T. kőningi*
5. Soil infusion (1 cc. of 1:5 soil suspension).

At definite incubation periods, duplicate flasks from each treatment were removed and their contents analyzed in a moist state by both the pipette and the slope methods. The residual material from each flask was dried at 50°C. for 24 hours, sieved through a 3-mm. screen, and again analyzed by both methods.

The results presented in tables 1 and 2 show that the various microbes had a definite effect upon the physical properties of the sand-bentonite mixture. This was due to a large extent to the holding together of the particles by the

extensive fungus mycelium or by the sticky substances produced by the bacteria. When the contents of the *A. niger* and *R. nigricans* flasks were ex-

TABLE 1

Binding effects of microorganisms upon the fine particles in sand-bentonite mixtures, with sucrose as energy source*

METHOD USED.....	PIPETTE					SLOPE				
State of material.....	Moist			Dry		Moist			Dry	
Incubation.....days	23		55	23	55	23		55	23	55
Inoculation	Un-bound material	Bound	Bound	Bound	Bound	Un-bound material	Bound	Bound	Bound	Bound
	gm.	per cent	per cent	per cent	per cent	gm.	per cent	per cent	per cent	per cent
Control.....	2.02	0	0	0	0	1.22	0	0	0	0
<i>R. nigricans</i>	2.00	2	0	3	5	1.05	18	15	36	33
<i>A. niger</i>	1.82	10	7	25	14	0.50	58	53	72	70
<i>Az. indicum</i>	1.66	18	27	26	25	0.60	50	61	60	75
<i>Ps. fluorescens</i>	2.33	-15	15	10	6	0.80	33	42	36	54
Soil infusion.....	1.70	17	9	17	0	0.66	41	33	45	41

* Unbound material calculated on basis of 50 gm. of dry material; bound material, on basis of control as 100 per cent unbound.

TABLE 2

Binding effects of microorganisms upon the fine particles in sand-bentonite mixture, with cellulose as energy source*

METHOD USED.....	PIPETTE					SLOPE				
State of material.....	Moist			Dry		Moist			Dry	
Incubation.....days	23		55	23	55	23		55	23	55
Inoculation	Un-bound material	Bound	Bound	Bound	Bound	Un-bound material	Bound	Bound	Bound	Bound
	gm.	per cent	per cent	per cent	per cent	gm.	per cent	per cent	per cent	per cent
Control.....	1.72	0	0	0	0	0.95	0	0	0	0
<i>T. koningi</i>	1.71	0	18	0	10	0.78	19	26	41	48
<i>Trichoderma</i> + <i>A. fumigatus</i>	1.88	-9	14	3	14	0.76	21	21	49	61
<i>Trichoderma</i> + <i>Penicillium</i>	1.74	0	21	6	27	0.78	19	24	45	52
Soil infusion.....	1.46	11	41	14	51	0.56	42	46	49	83

* Unbound material calculated on basis of 50 gm. of dry material; bound material, on basis of control as 100 per cent unbound.

amined it was noted that the whole mass was tightly held together by the mycelial network of the organisms. The contents of the flasks inoculated with *Az. indicum* were held together by a sticky mass produced by this organism.

The binding effect varied with the organism as well as with the method used for estimating this effect.

In the sugar series, the two fungi grew very quickly and produced the highest binding effect after 10 days' incubation, that of *A. niger* being much greater than that of *R. nigricans*. When measured by the pipette method, the binding effects of the fungi had decreased appreciably at the end of 55 days; by the slope method, however, the less drastic of the two procedures, the effects were still present to a considerable degree even at that time.

Azotobacter indicum grew more slowly than the fungi, but after 23 days its binding effect was almost as great as that of *A. niger*, and after 55 days its effect was even greater. *Ps. fluorescens* brought about the greatest binding of the fine particles after 55 days' incubation; however, this binding was not so stable as that of *Az. indicum*, since it did not show up so well under the more drastic treatment of the pipette method. Although the soil infusion did not produce as great a binding effect as *A. niger* or *Az. indicum*, its effect reached a maximum after 23 days' incubation, when it began to decrease.

In the cellulose flasks, the organisms grew much more slowly than those in the sucrose flasks, but they also brought about a marked binding of some of the particles in the sand-bentonite mixture. The effect was greater in 55 days than in 23 and greatest in the mixture receiving the soil infusion.

The reaction of the media did not change appreciably except in the case of *A. niger*, where it decreased to pH 5.6 in 23 days and then increased again. The reaction of the control flasks was approximately neutral. As the analyses of the duplicate flasks checked very well, only averages are reported.

Sand-clay mixtures

Two mixtures of sand and clay were employed, one consisting of 20 per cent clay and 80 per cent sand, and the other of 40 per cent clay and 60 per cent sand. The first mixture had a moisture-holding capacity of 36 per cent; and the second, 56 per cent. The sand was similar to the one used for the sand-bentonite experiment, and the clay was a light colored river clay with a moisture-holding capacity of approximately 80 per cent. Salt solutions similar to those employed with the sand-bentonite mixtures were added. Soil infusion was the only inoculum, except in one instance.

The flasks with contents were sterilized, inoculated, and incubated at 28°C. After 10 and 21 days, in the case of the flasks containing sucrose and no carbohydrate, and after 20 and 40 days, in the case of the flasks containing cellulose, duplicate portions from each treatment were removed and analyzed in the same manner as the sand-bentonite mixtures. The results are given in tables 3 and 4.

The microorganisms again produced a marked binding of the inorganic soil particles. The maximum effect occurred in 21 days in the sugar-treated soils and in 40 days in the soils receiving cellulose; the binding was greater in the moist material than in the dry. In the first series, *A. niger* produced results

TABLE 3
Binding effect of microorganisms in sand-clay mixtures*
80 per cent sand + 20 per cent clay

METHOD USED.....		PIPETTE						SLOPE					
State of material.....		Moist			Dry			Moist			Dry		
Incubation..... days		10	21	10	21			10	21	10	21		
Energy source	Inoculation	Unbound material	Bound	Bound	Bound	Bound	pH	Unbound material	Bound	Bound	Bound	Bound	pH
		gm.	per cent	per cent	per cent	per cent		gm.	per cent	per cent	per cent	per cent	
No carbohydrate.....	Control	8.22	0	0	0	0	6.7	4.22	0	0	0	0	6.7
	Soil infusion	8.22	0	3	1	1	6.6	3.82	9	20	10	13	6.6
Sucrose.....	Control	7.53	8	8	8	5	6.7	4.10	3	9	2	0	6.7
	Soil infusion	4.47	45	46	28	33	6.2	0.81	81	94	83	88	6.2
	<i>A. niger</i>	4.03	50	47	28	26	6.2	0.84	80	93	76	81	6.2
Cellulose.....	Control	8.06†	0	0†	0†	0†	6.3	1.50†	0	0†	0†	0†	6.3
	Soil infusion	6.75‡	16	10‡	8†	9†	6.4	1.03†	31	14‡	1†	39‡	6.4

* Unbound material calculated on basis of 50 gm. of dry material; bound material, on basis of control as 100 per cent unbound.

† Incubation, 20 days.

‡ Incubation, 40 days.

TABLE 4
Binding effect of microorganisms in sand-clay mixtures*
60 per cent sand + 40 per cent clay

METHOD USED.....		PIPETTE						SLOPE					
State of material.....		Moist			Dry			Moist			Dry		
Incubation..... days		10	21	10	21			10	21	10	21		
Energy source	Inoculation	Unbound material	Bound	Bound	Bound	Bound	pH	Unbound material	Bound	Bound	Bound	Bound	pH
		gm.	per cent	per cent	per cent	per cent		gm.	per cent	per cent	per cent	per cent	
No carbohydrate.....	Control	11.20	0	0	0	0	6.5	3.20	0	0	0	0	6.5
	Soil infusion	11.15	0	7	0	0	6.5	3.14	2	0	2	0	6.5
Sucrose.....	Control	11.37	0	0	2	0	6.4	2.61	18	11	6	-11	6.4
	Soil infusion	6.87	39	44	22	31	6.2	0.72	77	94	68	73	6.2
Cellulose.....	Control	11.67†	0	0†	0†	0†	6.1	0.52†	0	0†	0†	0†	6.1
	Soil infusion	10.17‡	13	20‡	5†	13†	6.2	0.22†	58	42‡	12‡	32‡	6.2

* Unbound material calculated on basis of 50 gm. of dry material; bound material, on basis of control as 100 per cent unbound.

† Incubation, 20 days.

‡ Incubation, 40 days.

similar to those obtained with the soil infusion. With no energy source, the inoculation gave very little, if any, binding effect. Because of the more abundant growth on sucrose, the organisms using this source of energy brought about greater binding than did those using cellulose.

The results of the determinations by the slope method reveal the fact that cellulose alone (in the control flasks) reduced the loss of the finer particles; part of the reduction may have been due to a lowering of the pH value, as will be shown later. The greater amount of clay in the second series of flasks seems to act as a binder itself. In a moist condition, more clay was washed out from the control mixture with the lower clay content than with the higher. In a dry condition about the same amount was washed out from both mixtures.

Although the added salt solutions contained a considerable amount of buffer, the pH was lowered in some instances as a result of growth of the organisms. When the pH was reduced to about 6.2, the clay particles appeared to be flocculated. For this reason, it was decided to determine the effect of reaction in binding the inorganic particles as measured by the methods used in this investigation. Accordingly, the salt solutions were adjusted to four different pH values and added to mixtures of 20 per cent clay and 80 per cent sand. The mixtures were sterilized, dried at 50°C., and screened, and duplicate samples from each lot were analyzed by the pipette and slope methods. The following results represent unbound material in the original 50-gm. samples:

pH	PIPETTE METHOD	SLOPE METHOD
	gm.	gm.
6.8	9.24	2.30
6.4	9.08	2.08
5.9	9.06	1.79
5.4	9.30	1.67

The reaction of the medium appears to have little effect upon the results obtained by the pipette method; but the results of the slope method are markedly affected, the amount of material washed out decreasing with a lowering in pH value. This would indicate that part of the binding effect found by the slope method with the material of lower pH values may be due to reaction change.

In order to determine the accuracy of the results obtained by the methods used in these studies, a number of samples of dried sand-clay mixtures were treated in the same manner as the "no-carbohydrate control flasks" and analyzed by both methods. The following results represent grams of unbound material in 50 gm. of the mixture:

PIPETTE METHOD			SLOPE METHOD		
9.065	9.220	9.370	2.020	2.205	2.610
9.150	9.255	9.395	2.100	2.435	2.685
9.180	9.285	9.410	2.125	2.435	
9.190	9.310		2.175	2.440	

A statistical examination of the results shows a standard deviation of ± 0.1 and a probable error of 0.067, for the pipette method, and a standard deviation of ± 0.21 and a probable error of 0.144 for the slope method. The pipette method is thus found to be more accurate than the slope method; the latter, however, may still give indications of relative binding effects of inorganic particles due to the growth of microorganisms.

Soil studies—sugar as source of energy

In order to determine the binding and aggregating effects of microorganisms on natural soil, two soils were selected, Bermudian clay loam and Collington sandy loam, with moisture-holding capacities of 39 and 50 per cent, respectively. The heavier soil is made up of material which has been eroded away from Penn soils. Since it was desirable to use a soil with a low organic matter content, only the subsoil was collected. The organic carbon content of the clay soil was 0.336 per cent, and that of the sandy soil 0.205. Both soils were air dried and sieved through a 2-mm. screen.

To 200-gm. portions of soil, placed in a series of flasks, the following salt solution was added, 40 cc. for the clay soil and 28 cc. for the sandy soil:

$(\text{NH}_4)_2\text{HPO}_4$	6.0 gm.
K_2HPO_4	5.0 gm.
H_2O to make.....	1000 cc.

Two per cent sucrose was used as an energy source. The sterilized flasks were divided into groups of six; one set from each soil type was inoculated as follows:

1. Control
2. *Az. indicum*
3. *Ps. fluorescens*
4. *A. niger*
5. *R. nigricans*
6. Soil infusion

Duplicate flasks from each soil and each treatment were analyzed, by both the pipette and slope methods, after 10, 22, and 50 days' incubation at 28°C. The methods were now somewhat modified. A 500-cc. graduate was used in place of a 250-cc. one in the pipette method, and 25-cc. aliquots were withdrawn for analysis. With the pipette method, the unbound soil, or the soil particles remaining in suspension after 1 minute, consisted approximately of the silt and clay particles. For each determination, 25-gm. portions of the heavy soil and 50-gm. portions of the sandy soil were used. With the slope method, 50-cc. instead of 200-cc. aliquots were withdrawn from the water containing the washed out soil particles.

The results of these studies are reported in tables 5 and 6. They show that the various microbes brought about a marked binding and aggregation of the soil particles, an effect much greater than that in the artificial soils.

In the clay loam soil, the fungi and the mixed soil population were more effective binding agents than the bacteria. *Ps. fluorescens* brought about very

little, if any, binding and even exerted a dispersing effect in the moist soil after 10 and 22 days' incubation. Because of the low pH value of the clay loam, *Ps. fluorescens* grew very slowly. *Azotobacter indicum* produced an effect intermediate between the two extremes. After 10 and 22 days, the

TABLE 5
Binding effects of microorganisms in clay loam soil

METHOD USED.....	PIPETTE						SLOPE					
State of material.....	Moist			Dry			Moist			Dry		
Incubation.....days	22		50	22		50	22		50	22		50
Inoculation	Un-bound material	Bound	Bound	Bound	Bound	pH	Un-bound material	Bound	Bound	Bound	Bound	pH
	gm.	per cent	per cent	per cent	per cent		gm.	per cent	per cent	per cent	per cent	
Control.....	18.0	0	0	0	0	4.5	3.03	0	0	0	0	4.5
<i>Az. indicum</i>	16.8	7	23	22	21	4.4	2.09	31	54	56	45	4.4
<i>Ps. fluorescens</i>	18.7	—3	4	2	3	4.4	3.32	—9	3	37	20	4.4
<i>A. niger</i>	12.6	30	54	45	46	4.3	0.72	76	87	79	76	4.3
<i>R. nigricans</i>	13.8	23	47	41	33	4.3	0.70	77	89	49	40	4.3
Soil infusion.....	13.9	23	64	37	46	4.6	0.54	82	93	47	73	4.6

TABLE 6
Binding effects of microorganisms in sandy loam soil

METHOD USED.....	PIPETTE						SLOPE					
State of soil.....	Moist			Dry			Moist			Dry		
Incubation.....days	22		50	22		50	22		50	22		50
Inoculation	Un-bound material	Bound	Bound	Bound	Bound	pH	Un-bound material	Bound	Bound	Bound	Bound	pH
	gm.	per cent	per cent	per cent	per cent		gm.	per cent	per cent	per cent	per cent	
Control.....	5.20	0	0	0	0	6.9	0.98	0	0	0	0	6.9
<i>Az. indicum</i>	3.76	28	29	65	62	6.7	0.66	33	44	81	59	6.7
<i>Ps. fluorescens</i>	4.91	6	14	31	30	6.9	0.83	15	31	41	54	6.9
<i>A. niger</i>	3.44	34	54	47	55	4.7	0.22	78	89	78	75	4.7
<i>R. nigricans</i>	2.66	49	58	32	42	4.7	0.11	89	91	64	62	4.7
Soil infusion.....	2.62	50	43	72	71	6.7	0.12	88	82	96	81	6.7

binding brought about by the fungi, *Az. indicum*, and the mixed soil population was greater in the dry soil; but after 50 days, it was greater in the moist soil. With the other organisms no great difference could be detected in the amount of bound soil in the moist and dry state.

The sandy soil gave generally similar results to the clay loam soil. *Azoto-*

bacter indicum, however, produced approximately as great a binding effect as the fungi. *Pseudomonas fluorescens* had a small aggregating effect, which was increased after the soil had been dried. In the moist state, *R. nigricans* brought about the greatest binding action in the sandy soil, but when the soil was dried the mixed soil population proved to be more effective. The mixed population caused 71 per cent binding in 50 days, as shown by the pipette procedure. The differences in the behavior of the soil in a moist and a dry state were probably due to the type of growth produced by the different organisms. *Rhizopus nigricans* made an extensive mycelial growth throughout the whole mass of sandy soil, the particles were held tightly together, and the mass felt spongy. As a result of drying and sieving, the mycelial network was broken up, resulting in a lowering of the binding effect. The soil inoculated with *A. niger* or with the mixed population tended to fall apart into a more or less granular state. Upon drying, some of the colloidal organic material resulting from growth of the organisms became irreversible, which would tend to stabilize the granules or bind the finer soil particles more tightly together.

Although the pipette method was shown to be more accurate than the slope method, both methods gave similar results. The relative amount of material bound, in accordance with the slope method, however, was greater than that bound according to the pipette method. This was due to the more drastic treatment of the soil in the latter method. It is also to be noted that the duplicates for the slope method checked much better in the sandy soil than in the clay loam soil.

During incubation of the flasks, the moisture content of the soil gradually decreased. This was accompanied by a decrease in the amount of unbound material in the control flasks, pointing to the fact that the moisture content of the soil affects the aggregation of the finer soil particles. This phenomenon was also noted by Cholodny (6), who found the effect to be very marked. After the soils were dried at 50°C., the influence of the original moisture content was no longer apparent. The amount of unbound material in the dry control soils remained virtually the same throughout the experiment. The rapid drying of the soils at 50°C. increased dispersion, however, because the weight of unbound material in the dry control soils was consistently more than that in the moist control soils, when measured by the pipette method.

Soil studies—use of complex organic materials

When sucrose is used as an energy source, microbial growth takes place quickly and a large amount of cell material is formed. The binding effects produced on the soil under such conditions are largely due to the cell material synthesized by the organisms. The sucrose itself rapidly disappears and has little, if any, aggregating effect upon the soil constituents. Complex organic materials, such as alfalfa, manure, and peat, contain substances which are

less readily available to the soil organisms. When these materials are decomposed, the binding effect on the soil may be a result not only of the synthesized cell substance but also of the organic constituents of the material undergoing decomposition. The easily available organic compounds are utilized by the organisms for energy and for cell synthesis, whereas the more resistant materials are left free to undergo various physical and chemical interactions with the inorganic soil colloids, which may result in increased aggregation of the soil as a whole.

TABLE 7

Binding effect of microorganisms upon soil, with complex organic materials as energy sources
Soil not sterilized

STATE OF SOIL.....	MOIST				DRY				
Incubation.....days	20		50		20		50		
Treatment*	Un-bound material	Bound	Un-bound material	Bound	Un-bound material	Bound	Un-bound material	Bound	pH
	gm.	per cent	gm.	per cent	gm.	per cent	gm.	per cent	
Control.....	6.47	0	5.93	0	6.45	0	6.10	0	6.7
Alfalfa.....	3.65	44	3.84	36	2.80	57	2.35	62	7.2
Manure.....	5.42	17	4.62	23	5.24	19	4.54	26	7.0
Peat.....	6.43	0	5.57	6	5.91	8	5.94	3	6.3

* Organic materials used in 2 per cent concentration.

In order to study the effect of complex organic materials on the binding and the aggregation of the soil particles, 200-gm. portions of Collington sandy loam were treated as follows:

1. Control
2. Alfalfa, 2 per cent
3. Manure, 2 per cent
4. Lowmoor peat,² 2 per cent

The organic materials were dried and finely ground before they were used. The mixtures were placed in 500-cc. Erlenmeyer flasks, and the moisture content was adjusted to 55 per cent saturation. Each flask was inoculated with 1 cc. of a soil suspension, and the flasks were placed in the incubator at 28°C.

A second series of flasks was prepared in a similar manner; in this case the flasks were sterilized, and one half were inoculated with a spore suspension of *A. niger* and the other with a suspension of a fertile soil. Duplicate flasks from each treatment and from both series were analyzed in the moist and dry state by the pipette method, after 20 and 50 days' incubation.

The results, presented in tables 7 and 8, show that in Collington sandy loam the greatest binding effect was produced by the addition of alfalfa. The bind-

² New Jersey cultivated peat known as Hyper-Humus.

ing was very marked, being as great as 60 or 70 per cent in the dried and screened soil after 50 days' incubation. The lowmoor peat produced only a slight aggregation. The effect of the manure treatment was intermediate. Alfalfa contains the greatest amount of readily available organic material for the organisms and therefore allows much more abundant growth of the microorganisms. This factor and the chemical nature of the alfalfa were probably responsible for the marked aggregating effect.

The amount of unbound material was approximately the same in both the sterilized and unsterilized control and alfalfa-treated soils. The manure and peat treatments, on the other hand, exerted the greater aggregating effect in the sterilized soil. It is possible that the sterilization process made some of

TABLE 8

Binding effect of microorganisms upon soil, with complex organic materials as energy source
Soil sterilized

STATE OF SOIL.....		MOIST				DRY				
Incubation.....days		20		50		20		50		
Inoculation	Treatment*	Un-bound material	Bound	Un-bound material	Bound	Un-bound material	Bound	Un-bound material	Bound	pH
		gm.	per cent	gm.	per cent	gm.	per cent	gm.	per cent	
<i>A. niger</i>	Control	6.79	0	5.78	0	6.59	0	6.39	0	6.7
	Alfalfa	4.40	36	3.57	39	3.40	49	2.92	54	6.5
	Manure	5.45	20	4.02	30	5.21	21	4.87	24	7.1
	Peat	6.08	10	5.30	8	6.13	7	5.73	10	6.5
Soil suspension	Control	6.46	0	5.69	0	6.34	0	6.32	0	6.7
	Alfalfa	3.91	39	2.41	58	2.72	58	1.90	70	7.1
	Manure	4.61	22	3.27	43	4.61	28	4.00	37	7.0
	Peat	6.02	0	4.21	26	5.95	6	5.30	17	6.5

* Organic materials used in 2 per cent concentration.

the constituents of the manure and peat more available to the soil organisms. *Aspergillus niger* produced the greatest aggregating effect with alfalfa as an energy source, an intermediate effect with manure, and only a slight effect with peat. The binding action brought about by *A. niger*, however, was not so great as that of the mixed soil population. There was a considerable increase in aggregation upon drying of the alfalfa-treated soil. In the other cases the aggregation was approximately the same in both the moist and dry states. The variation in chemical composition of the substances undergoing decomposition was probably responsible for these differences.

An attempt was next made to determine the effect of lime on the binding action of organic matter undergoing rapid decomposition. For this purpose,

Bermudian clay loam, having a pH of approximately 4.6, was used. It was treated, in 200-gm. portions, as follows:

1. Control
2. CaCO_3 , 0.1 per cent
3. CaCO_3 , 0.5 per cent
4. Alfalfa, 2 per cent
5. Alfalfa, 2 per cent + CaCO_3 , 0.1 per cent
6. Alfalfa, 2 per cent + CaCO_3 , 0.5 per cent

After adjustment of the moisture content and inoculation with 1 cc. of a soil suspension, the flasks were incubated at 28°C . At 20 and 45 days they were analyzed in duplicate by the pipette method.

TABLE 9
Binding effect of lime and alfalfa upon soil

STATE OF SOIL.....	MOIST				DRY				
Incubation.....days	20		45		20		45		
Treatment	Un-bound material	Bound	Un-bound material	Bound	Un-bound material	Bound	Un-bound material	Bound	pH
	gm.	per cent	gm.	per cent	gm.	per cent	gm.	per cent	
Control.....	20.97	0	17.32	0	21.40	0	20.26	0	4.6
CaCO_3 , 0.1%.....	20.78	0	16.60	4	22.48	—4	19.81	2	5.7
CaCO_3 , 0.5%.....	21.00	0	16.34	5	23.54	—10	20.64	0	7.1
Alfalfa, 2%.....	14.88	29	14.68	16	15.50	28	14.00	31	5.0
Alfalfa, 2% + CaCO_3 , 0.1%.....	14.69	30	11.39	35	16.04	26	12.78	37	5.8
Alfalfa, 2% + CaCO_3 , 0.5%.....	14.55	31	11.16	36	15.02	30	11.36	44	7.1

The results, presented in table 9, indicate that lime alone had no aggregating effect upon the soil. The alfalfa alone produced a considerable binding. After 20 days, the lime together with the alfalfa exerted no additional effect over that of alfalfa alone; but after 45 days, an additional binding effect occurred. This experiment is only preliminary in nature and does not warrant the drawing of broad conclusions; however, it indicates that lime together with organic matter may in time increase the aggregation of the soil. More detailed experiments on this question are now in progress.

Effect of different sources of organic matter

A study was next made of the effects of different types of organic matter, particularly at different stages of decomposition, on those physical properties of the soil that have a bearing upon erosion. Sassafras clay loam, from which the top 6 inches were discarded, was used in these experiments. The

soil was air dried; screened; and placed, in 20-kgm. portions on a dry basis, in five large metal containers treated as follows:

1. Control
2. Ground alfalfa, 1 per cent
3. Ground wheat straw, 1 per cent + ammonium phosphate, 0.05 per cent
4. Peat moss, 1 per cent
5. Fresh horse manure, 1 per cent

The water content of the soil was brought to 50 per cent saturation, and the containers were incubated at 28°C., the optimum water content being maintained by frequent additions of water. At the end of 1, 2, 4, 8, and 12 weeks, the soils were analyzed for organic matter, percolation rate, dispersion ratio and moisture-holding capacity. The organic carbon was determined by a modification of Schollenberger's method (28), and the organic matter was calculated by the use of the factor 1.72. The percolation rate was measured

TABLE 10

Rate of decomposition of different organic materials and their effect upon the physical properties and moisture-holding capacity of a Sassafras soil

PERIOD OF INCUBATION...weeks	1	4	12	1	4	12	1	4	12	1	4	12
Treatment of soil	Organic matter left			Percolation rate			Dispersion ratio			Water-holding capacity		
	per cent*	per cent*	per cent*	cc./hr.	cc./hr.	cc./hr.				per cent	per cent	per cent
Control.....	1.72	1.72	1.67	105	150	110	47	47	51	63	61	58
Alfalfa.....	2.38	2.24	1.90	500	490	500	38	28	33	65	68	68
Straw.....	2.40	2.24	1.87	420	300	190	39	36	48	66	67	61
Peat.....	2.57	2.55	2.27	700	600	600	46	37	46	69	70	68
Manure.....	2.50	2.32	2.13	450	360	280	41	41	44	64	67	65

* Of dry soil.

according to the method of Slater (24) using a cylinder 4 inches in diameter filled to a depth of 7 inches. For the dispersion ratio the method of Middleton (15) was used, except that the Bouyoucos hydrometer was employed in place of a pipette for determining the amount of silt and clay in suspension. The moisture-holding capacity was determined by the Hilgard method. All the analyses were made in duplicate.

The results of this experiment are reported in table 10. The addition of organic matter was found to have a marked effect upon the physical properties of the soil. All four forms of organic matter used increased the percolation rate, peat and alfalfa being the most effective. The alfalfa and straw decomposed at a rapid rate, the manure somewhat more slowly, and the peat to only a limited extent. As the straw and manure underwent decomposition, the percolation rate decreased; in the alfalfa-treated soil, however, the percolation rate remained high, even at the end of 3 months.

The degree of soil dispersion was expressed by the ratio of the amount of silt and clay when determined without dispersion to that obtained by mechanical analysis. The lower the dispersion ratio, the greater was the number of water-stable aggregates larger than the silt and clay particles. In this investigation, the addition of alfalfa, straw, and manure brought about a marked increase in the number of aggregates, as measured by the dispersion ratio; alfalfa produced the greatest effect. Peat caused only a small increase in aggregation as compared with the other organic materials. After 3 months, the increase in aggregation in relation to the control had almost disappeared in the peat- and in the straw-treated soil; it was still apparent in the soil to which manure had been added and was very marked in the soil receiving alfalfa.

All four types of organic matter increased the moisture-holding capacity of the soil.

DISCUSSION

A study of the binding effect on soil particles produced by pure and mixed cultures of microorganisms in artificial and natural soils supplied with readily decomposable organic materials brought out emphatically the fact that microorganisms play a highly important role in conserving the soil from removal by leaching and erosion. Both in artificial soils—sand-bentonite and sand-clay mixtures—and in natural heavy and light soils, it was possible to show that the growth of microorganisms led to a binding and aggregation of the soil particles. The extent of binding was found to be dependent upon the nature of the microorganisms, the amount of growth produced, and the nature of the substrate. *Rhizopus nigricans* and *Aspergillus niger* made good growth in the sand-bentonite mixture, but the latter produced a much greater binding effect. Some of the binding brought about by the fungi, in particular by *R. nigricans*, was due to a mechanical holding together of the soil by an extensive mycelial network produced by the organisms. After the mycelial network was broken up by screening, the binding effect of *R. nigricans* on the sandy soil was considerably reduced. Besides a mechanical binding of the soil particles, the organisms brought about an aggregation due to the synthesized organic compounds. The extent of binding remaining even after the soils were dried and screened indicates that this effect was very marked. In water, the control soils rapidly slaked into a sticky mass, but the soils in which growth of microorganisms had occurred remained in a more or less granular condition.

Azotobacter indicum grew much better in the sandy soil than in the heavy clay soil. It produced a great deal of slimy material which cemented the soil particles together; drying increased its effectiveness. *Pseudomonas fluorescens* was consistently the poorest agent in this respect.

A much larger amount of soil was bound in the heavy clay soil and in the sand-clay mixtures than in the light sandy soil and in the sand-bentonite mixtures; the percentage of material bound, however, did not differ greatly.

The extent and duration of the binding effect and the time required for it to reach a maximum was found to depend not only on the organisms involved but also on the type of organic material used. In the sand-bentonite and sand-clay mixtures, it was shown that much more rapid growth was made with sugar as a source of energy than with cellulose; consequently, the binding effects appeared much sooner in the sugar-treated mixtures. When complex organic materials were used as sources of energy, it was found that the more readily a material can decompose, the greater is the binding action. The effect of lime was of particular interest in this connection: lime alone had no appreciable binding effect upon an acid clay soil, but in the presence of decomposing organic matter it increased the binding action of the latter, especially during the advanced stages of decomposition.

The foregoing results were duplicated with a different soil, a greater variety of materials, and different methods of measuring the physical soil properties. This experiment, as well, brought out the fact that greater beneficial effects of organic matter on the state of aggregation of a soil are produced more quickly by materials less resistant to microbial decomposition. The duration of the beneficial effects seems to be dependent not only upon the rapidity of decomposition but also upon the chemical nature of the organic matter. Both alfalfa and straw decomposed rather rapidly, but the influence of the straw upon the physical properties of the soil tended to disappear much earlier than did that of the alfalfa.

SUMMARY

The results of investigations on the role of microorganisms in bringing about soil aggregation have been reported. This was shown to have an important bearing upon the problem of soil erosion.

Two artificial soils—sand-bentonite and sand-clay mixtures—and two natural soils—Bermudian clay loam and Collington sandy loam—were used. To these were added various carbohydrates and plant residues. Pure and mixed cultures of microorganisms were used as inoculants. After incubation, two methods, designated as the pipette and slope methods, were used for measuring the binding effect of the activity of the microorganisms upon the finer soil particles.

The action of microorganisms was found to result in a marked binding and aggregation of the soil particles; the extent of the binding depended on the organisms concerned and the nature of the organic material added.

The results of the addition of several types of organic matter on certain physical properties of a natural soil were also reported. It was found that the effect of these materials depended entirely on their nature and the rapidity of their decomposition. The more rapidly a plant material decomposes, the greater will be its binding action upon the finer soil particles.

The effect of lime in bringing about an aggregation of the soil constituents appears to be associated largely with its effect upon the action of the organic materials.

REFERENCES

- (1) ANNE, P. 1935 Les fumures organiques, l'état calcique et la dispersion de l'argile dans les limons. *Ann. Agron.* 5: 781-785.
- (2) BAVER, L. D. 1935 Aggregation of desert soils. *Amer. Soil Survey Assoc. Bul.* 16: 54-55.
- (3) BAVER, L. D. 1935 Factors contributing to genesis of soil microstructure. *Amer. Soil Survey Assoc. Bul.* 16: 55-56.
- (4) BAVER, L. D., AND RHOADES, H. F. 1932 Aggregate analysis as an aid in the study of soil structure relationships. *Jour. Amer. Soc. Agron.* 24: 900-906.
- (5) BROWNING, G. M. 1937 Changes in erodibility of soils brought about by application of organic matter. *Soil Sci. Soc. Amer. Proc.* 2: 85-96.
- (6) CHOLODNY, L. The relation of degree of dispersion to moisture content of soil. *Proc. Soil Sci. Soc. Amer.* 1939 (in press).
- (7) DEMOLON, A., AND HENIN, S. 1932 Recherches sur la structure des limons et la synthèse des agrégats. *Soil Res.* 3: 1-9.
- (8) GELTZER, F. U. 1937 The role of organic matter in the formation of soil structure. *Chemization Socialistic Agr. (U. S. S. R.)* 7: 45-61; 8: 53-63.
- (9) KANIVETZ, I. I., AND KORNEEVA, N. P. 1938 Importance of biochemical structure formers. *Pedology (Russian)* 32: 1429-1441.
- (10) LUNT, H. A. 1937 The effect of forest litter removal upon the structure of the mineral soil. *Soil Sci. Soc. Amer. Proc.* 1: 59.
- (11) LUTZ, J. F. 1934 The structure of soils as affecting soil erosion. *Amer. Soil Survey Assoc. Bul.* 15: 98-100.
- (12) LUTZ, J. F. 1935 The relation of soil erosion to certain inherent soil properties. *Soil Sci.* 40: 439-457.
- (13) MADHOK, M. R. 1937 Synthetic soil as a medium for the study of certain microbiological processes. *Soil Sci.* 44: 319-322.
- (14) METZGER, W. H., AND HIDE, J. C. 1938 Effect of certain crops and soil treatments on soil aggregation and the distribution of organic carbon in relation to aggregate size. *Jour. Amer. Soc. Agron.* 30: 833-843.
- (15) MIDDLETON, H. E. 1930 Properties of soils which influence soil erosion. *U. S. Dept. Agr. Tech. Bul.* 178: 1-16.
- (16) MUSGRAVE, G. W. 1935 The infiltration capacity of soils in relation to control of surface run-off and erosion. *Jour. Amer. Soc. Agron.* 27: 336.
- (17) MUSGRAVE, G. W., AND NORTON, R. A. 1937 Soil and water conservation investigations at the Soil Conservation Experiment Station, Missouri Valley, Loess Region, Clarinda, Iowa. *U. S. Dept. Agr. Tech. Bul.* 558.
- (18) MYERS, H. E. 1937 Physicochemical reactions between organic and inorganic soil colloids as related to aggregate formation. *Soil Sci.* 44: 331-357.
- (19) PASCHAL, A. H., BURKE, R., AND BAVER, L. D. 1935 Aggregation studies on Muskingum, Chester, and Lansdale silt loams. *Amer. Soil Survey Assoc. Bul.* 16: 44-45.
- (20) PEELE, T. C. 1937 Effect of lime and organic matter on erodibility of Cecil clay. *Proc. Soil Sci. Soc. Amer.* 2: 79-84.
- (21) PEELE, T. C. 1940 Microbial activity in relation to soil aggregation. *Jour. Amer. Soc. Agron.* 32: 204-212.
- (22) SHRIKHANDE, J. G. 1935-36 The production of mucus during decomposition of plant materials: I. The effect of environmental conditions. *Biochem. Jour.* 27: 1551-1562; 30: 1789-1794.
- (23) SIDERI, D. I. 1938 On the formation of structure in soil: IV, V. *Soil Sci.* 46: 129-136, 267-270.

- (24) SLATER, C. S., AND BYERS, H. G. 1931 A laboratory study of field percolation rates on soils. U. S. Dept. Agr. Tech. Bul. 232.
- (25) SMITH, J. B., BROWN, P. E., AND RUSSELL, J. A. 1937 The effect of organic matter on infiltration capacity of Clarion loam. *Jour. Amer. Soc. Agron.* 29: 521-525.
- (26) SOKOLOVSKY, A. N. 1930 The problem of soil structure. *Soviet Sect. Internatl. Soc. Soil Sci.* 1: 94-98.
- (27) STEPHENSON, R. E. 1918 The effects of organic matter on soil reaction. *Soil Sci.* 6: 413-439.
- (28) WAKSMAN, S. A. 1916 Soil fungi and their activities. *Soil Sci.* 11: 103-155.
- (29) WAKSMAN, S. A. 1938 Humus, ed. 2. Williams & Wilkins Co., Baltimore, Md.
- (30) WAKSMAN, S. A., AND MARTIN, J. P. 1939 The role of microorganisms in the conservation of the soil. *Science* (n.s.) 90: 304-305.

A RAPID-ACTION SOIL TUBE JACK¹

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In obtaining samples with the soil tube, the extraction of the tube from the ground is often the most strenuous part of the operation. In sampling to depths of 3 to 5 feet two men usually are able to pull the tube by direct lift with the hammer between them. There are, however, many occasions when it is desirable for one man to be able to obtain samples from depths of 3 or 4 feet. The older form of jack² will pull the tube but is not entirely satisfactory, particularly in heavy wet soils. On wet clay adobe soils the clamp and the outside of the tube become badly fouled with greasy clay, and it is difficult to make the clamp grip the tube without some slipping. Under some conditions, the clamp must be continually released and set by hand to insure gripping. The stroke of the jack is necessarily short because of the danger of kinking the tube close to the ground. In addition, it is a relatively expensive piece of equipment.

The device described here (fig. 1) was constructed to provide a simple, fast-acting jack, low in cost, for use in soil moisture sampling by the commercial orchardist, and in other cases where one man only is available for the work.

This jack has proved to be very efficient and, for sampling-tube lengths up to 5 or 6 feet, is more quickly and easily used than the older type. It has a relatively long stroke and is ready for use with no loose parts to clean or set up and adjust. The operator controls the gripping of the fulcrum block by lightly squeezing together the short control lever and the shaft or upright member of the jack. To lift the fulcrum for a new bite the pressure on the control rod is relaxed. Any tendency for the jack to throw a side thrust on the tube in uneven or wet ground is controlled by the operator's hand on the jack shaft. This form of jack, of course, has a definite limitation as to length of soil tube which can be used. A jack shaft 5 to 5½ feet long, suitable for

¹ The device herein described was developed in connection with work conducted cooperatively by the U. S. Bureau of Plant Industry and Oregon Agricultural Experiment Station at the Medford Branch Experiment Station. This report is published as Technical Paper No. 330 with the approval of the director of the Oregon Agricultural Experiment Station and the chief of the U. S. Bureau of Plant Industry.

² Taylor, C. A., and Blaney, H. F. 1929 An efficient soil tube jack. *Soil Sci.* 27: 351-353.

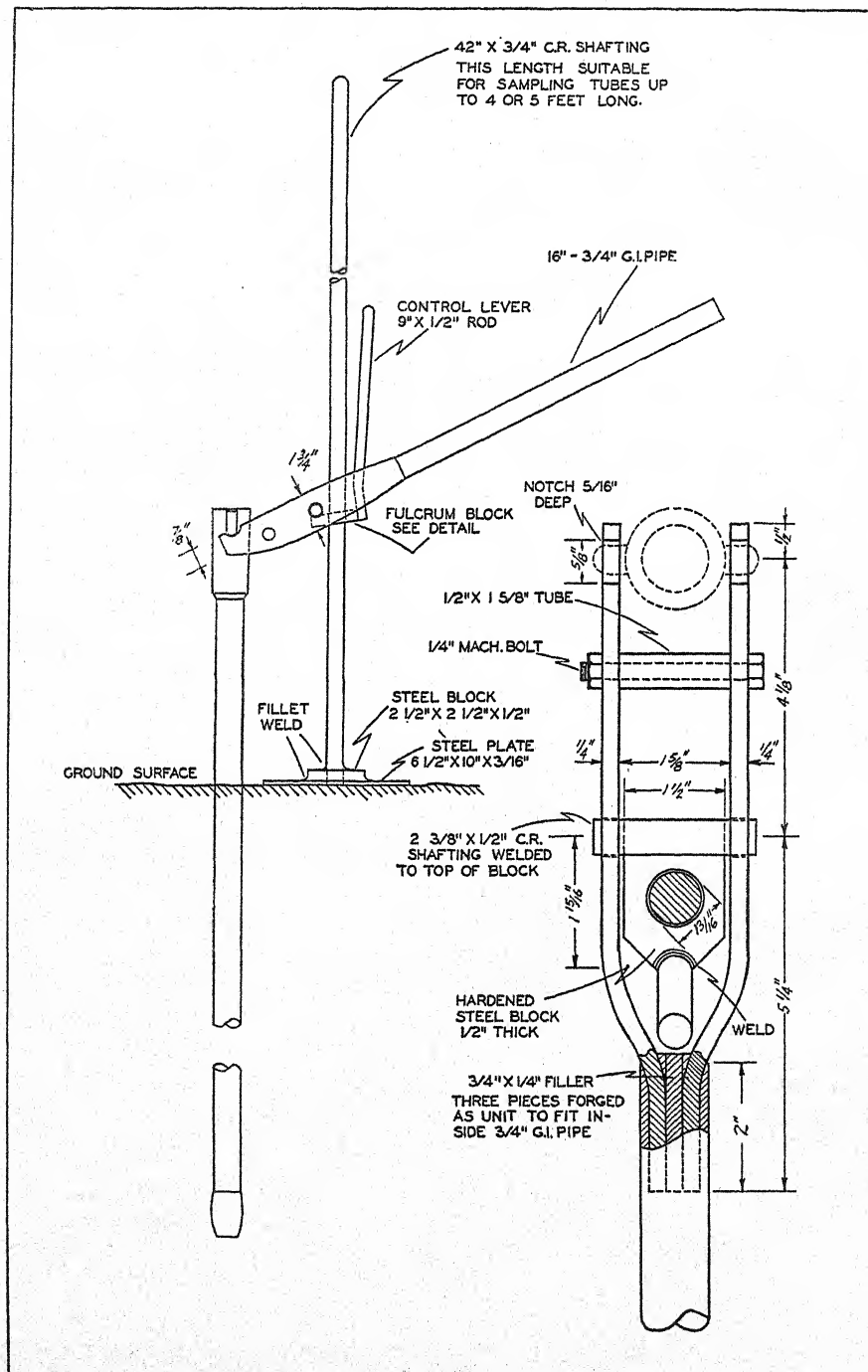
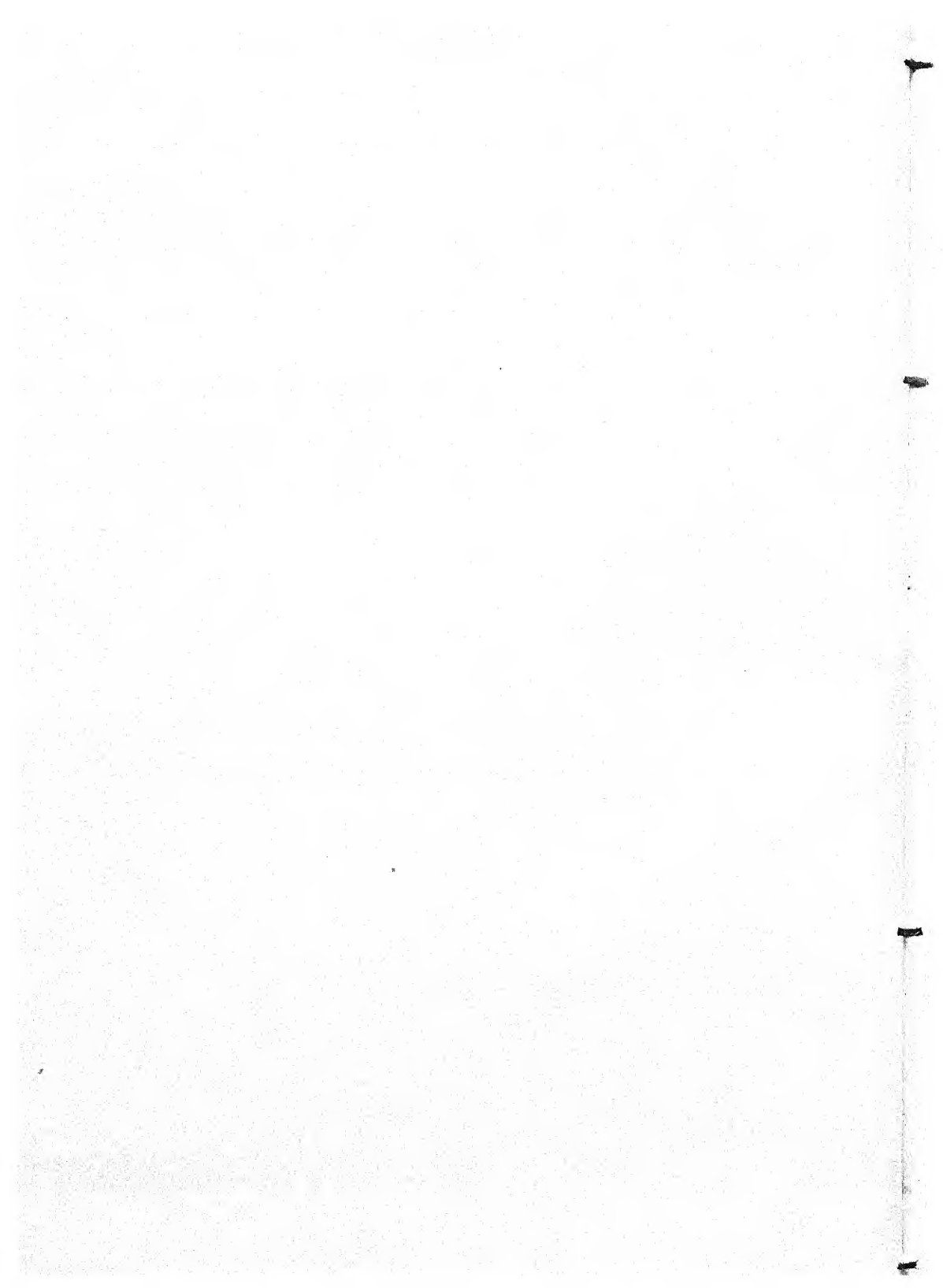


FIG. 1. CONSTRUCTION DETAILS OF THE SOIL TUBE JACK

tube lengths up to 6 feet, is about the maximum length that can be conveniently handled. The 42-inch shaft shown is satisfactory for tubes up to 4 or 5 feet long, since the last foot or so, usually, is easily pulled by hand. Where shorter tubes are used, the jack shaft can be shortened somewhat for lighter weight and convenience. A threaded extension of the shaft could be provided for longer tubes, or an additional longer shaft, complete with bottom plate, could be made for occasions when deeper sampling is undertaken.



ACCUMULATION OF ZINC ON SOIL UNDER LONG-PERSISTENT VEGETATION¹

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The finding of unusually large amounts of zinc in the topsoil of several widely separated localities of California where a heavy growth of trees or grass had been long undisturbed was reported recently (4). The zinc content was much higher than that in similar nearby soils where there had been no long accumulation of vegetable matter. It seemed probable that the accumulation of zinc was caused by action of the plants in bringing up from the subsoil much mineral matter. Then, when the fallen leaves and other debris decayed, the zinc and other heavy metals contained in them were fixed in the topsoil instead of going back to the subsoil from which they came. Unknown to the writer similar observations were reported by Goldschmidt in 1937 (1). To obtain some confirmation of the validity of this hypothesis the experiments reported in this paper were made.

EXPERIMENTAL

Materials and methods

Soil samples were taken under three kinds of trees, redwood, pine, and oak, in locations where it is probable that the ground cover and the soil itself had not been disturbed for many years. The layer of leaf material, in all stages of decay, varied from 2 to 6 inches deep on top of the soil. The soil samples represented depths of 0-1, 1-3, 3-5, 5-11, and 11-17 inches. At the same time samples of the vegetable matter were collected representing fresh green leaves, recently fallen unleached leaves, long-fallen probably leached leaves, and the much-decayed matter underneath resting on the surface of the soil. After being dried and ground, the samples were analyzed for zinc. The methods used for soil are described in previous papers (2, 3, 4). The leaf materials were burned, and total zinc in the ash was determined by the method previously described (2). Results are reported in table 1 for soils, in table 2 for the leaf materials. The figures plainly indicate that the decay of organic matter caused accumulation of zinc in the topsoil.

¹ Assistance in the preparation of these materials was furnished by the personnel of Works Progress Administration Official Project #465-03-3-587-B-10.

Corroborative experiments

These experiments were intended to simulate by laboratory work the long-time process of nature. Into filter tubes about 6 inches long were placed 100 gm. of a powdered soil, Yolo silty clay loam, which made a column about 3 inches deep. On top of the soil in three of the tubes was placed 2 gm. of the

TABLE 1
Available zinc in soils under large trees*

DEPTH	REDWOOD	PINE	OAK
<i>inches</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>
0-1	40.6	55.8	67.7
1-3	19.4	26.9	18.7
3-5	9.4	23.8	11.9
5-11	8.9	8.8	8.6
11-17	8.4	7.9	9.9

* The soils are similar physically.

TABLE 2
Total zinc in leaves of trees

LEAVES	REDWOOD	PINE	OAK
	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>
Fresh green.....	39	34	40
Recently fallen.....	53	37	92
Long fallen.....	83	43	70
Much decomposed.....	97	150	98

TABLE 3
Available zinc in soils after percolation with leaf extract

	ZINC IN LEAVES	VOLUME OF PERCOLATE	ZINC IN PERCOLATES	AVAILABLE ZINC IN SOILS		
				Top	Middle	Bottom
	<i>p.p.m.</i>	<i>cc.</i>	<i>gammas</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>
Soil only.....		250	10.5	4.5	4.5	4.2
Soil + redwood leaves....	39	200	7.7	5.5	4.5	4.5
Soil + pine leaves.....	34	250	11.9	5.0	4.8	3.9
Soil + oak leaves.....	40	175	10.5	5.3	5.3	4.8

dried, powdered fresh leaves of the three kinds of trees, redwood, pine, oak. One tube as a check, contained only soil without the leaf material. Distilled water was percolated through the leaf material and soil columns during 9 days. The volume of percolate from the tubes varied somewhat (see table 3), but the amount of zinc extracted was about 0.1 p.p.m. of the soil from each. The check tube percolate contained about the same amount of zinc as the percolate

from the tubes containing leaf material, which proves that none of the zinc leached out of the organic material had passed through the soil columns.

In a separate experiment 2 gm. of the dry powdered fresh green leaves were percolated with water during 3 days. The water extracted from redwood 19 gammas of zinc, from pine 22 gammas, from oak 33 gammas. In the leaching of the soil columns on which were placed the powdered leaf materials, if the same amounts of zinc were extracted from them in the presence of the soils and if all the zinc were retained by the soil, the zinc content of the soils would be increased. The increase of zinc in the soil from redwood would be 0.19 p.p.m., from pine 0.22 p.p.m. and from oak 0.33 p.p.m. Since all of this added zinc was retained by the soil there was no increase in zinc in the percolates over that from the soil alone. The figures in table 3 show that the available zinc in the top of the soil columns to which leaf material was added was appreciably increased over that in the soil alone.

DISCUSSION

Figures in table 3 show that the zinc extracted from the leaf material by the water accumulated in the top portions of the soil just as it does in nature under the trees. It is true, as would be expected, that the increase of zinc was small compared with that found in nature, where the accumulated organic matter was much thicker and the time of action was immensely longer.

The results of this experiment, as well as the results of analyses of the soils and leaf materials reported in tables 1 and 2, support the hypothesis that long-continued accumulation and decay of organic matter under trees or other heavy vegetation causes accumulations of considerable amounts of zinc and other mineral matter² which are easily fixed by the soil near its surface. This increase of zinc in the topsoil may be several times as much as the amount in the same soil which has not received the accumulations derived from mineralization of the fallen leaves during many years.

REFERENCES

- (1) GOLDSCHMIDT, V. M. 1937 The principles of distribution of chemical elements in minerals and rocks. *Jour. Chem. Soc.* part 1: 655-673.
- (2) HIBBARD, P. L. 1938 Estimation of copper, zinc and cobalt (with nickel) in soil extracts. *Indus. and Engin. Chem., Analyt. Ed.* 10: 615-618.
- (3) HIBBARD, P. L. 1940 Chemical status of zinc in the soil with methods of analysis. *Hilgardia* 13: 1-29.
- (4) HIBBARD, P. L. 1940 A soil zinc survey in California. *Soil Sci.* 49: 63-72.

² Other analyses, not here reported, show that in the topsoil under heavy accumulations of organic matter there are increased amounts of Cu, Pb, Ni, and Co.

LYSIMETER STUDIES: IV. MOVEMENT OF ANIONS THROUGH THE PROFILE OF A GRAY-BROWN PODZOLIC SOIL¹

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This paper presents summary data on the composition of the anions of leachings collected during 7 years from lysimeters of the funnel type.

In brief, the lysimeter consists of a funnel made of block tin, 30 cm. in diameter, 4 to 5 cm. deep, with nine or ten 2-mm. perforations in the center. It is filled with quartz pebbles and coarse sand, and its sharp edge, 3 to 5 mm., rests against the roof of a tunnel dug under each horizon of the soil profile.² The tunnel depth is about 50 cm. Thus, when the funnel is inserted in place, the distance from the end of the funnel nearest to the face of the soil profile is about 20 cm. Four-by-four blocks of wood are placed against the face of the profile, and boards are nailed on. The space between the soil profile face and the boards (about 10 cm.) is then filled with the soil material that came from the excavation of the pit. The funnels are located under the respective horizons about 100 cm. apart on a horizontal line. Block tin tubes connect each funnel to Erlenmeyer flasks where the leachings are caught.

The leachings are collected and analyzed after each rain. Determinations are made on anions, cations, pH, and conductivity. Data were published on the movement and translocation of the cations and of some of the anions for the first 2 years (2, 3, 4). At that time definite trends on the cations were noted, but the data on the anions were not clear-cut. As the analytical data accumulated, the picture of the anions also became clearer.

Of the data presented, only those on the A horizon can be considered to be quantitative. The reason for the difference in the quality of the data from the A and B horizons is that the movement of the water through the A horizon is vertical and through the B only partly so. The rate of movement of water varies with the soil horizon, being faster through the A horizon, because of the lighter texture and the more open structure. The heavier texture and more compact constitution of the B horizon will therefore cause water to accumulate at the junction of the A and B horizons. Under such circumstances the water begins to move horizontally along the B horizon, through which only a portion of it filters, most of it dropping through by way of chan-

¹ Journal Series paper of the New Jersey Agricultural Experiment Station, department of soil chemistry and bacteriology.

² To expose the profile a pit is dug, 120 cm. wide, 180 cm. deep, and 360 cm. long. The pit is completely enclosed.

nels: cracks produced by the drying out of the soil, root paths, and passageways and borings of earthworms, rodents, and other animals.

RESULTS

Chlorine

The data in table 1 show that about $4\frac{1}{2}$ pounds of Cl per acre filter through the A₁; about 2 pounds through the A₂³; and only 0.63 pound through the B₁ horizon.⁴

It is to be noted that of the Cl that reaches the A₂ horizon less than half passes through it. The rest is retained by this horizon, apparently for the use of plants and animals harboring in the soil. Only about one-third of the Cl that passes through the A₂ appears in the leachings of the B horizon. An unknown quantity of Cl passes through the B horizon by way of the aforementioned channels.

TABLE 1
Annual translocation of anions through the soil profile
Average of 7 years

HORIZON		ANNUAL VOLUME OF LEACHING PER FUNNEL	TOTAL SOLIDS	Cl	NITRATE N	S	P	SiO ₂
Designation	Depth							
	cm.	liters	lbs./A.	lbs./A.	lbs./A.	lbs./A.	lbs./A.	lbs./A.
A ₁	18	28.863	340.7	4.45	6.86	28.4	0.33	8.77
A ₂ (e)*	24	6.535	87.5	1.61	2.41	14.1	0.065	2.44
A ₂ (s)*	24	6.593	72.4	2.03	2.27	10.2	0.052	3.19
B ₁	16	1.887	38.7	0.63	0.30	2.08	0.016	1.61

* The letters "e" and "s" are the signs for the duplicate funnels in the A₂ horizon. The other funnels are not duplicated.

An interesting feature of the Cl data is the concentration differential in the leachings of the respective horizons. The Cl concentration is about the same in the leachings of the A₂ and B₁ horizons and lower in the leachings of the A₁ horizon.

The curve of the Cl in figure 1, showing the leachings of the A₁ horizon, brings out a striking characteristic: a regularity of successive peaks and dips, indicating that the composition of the rainfall varies from time to time with a definite regularity. The explanation for this regularity was puzzling for a

³ By averaging the data of the two funnels, the quantity percolating is found to be 1.82 pounds per acre. It is to be remembered that the figures on the leachings from the individual lysimeter funnels must be multiplied by 55,000 to convert the data to the acre basis, the diameter of the funnel being equal to 1/55,000 of an acre. The difference of 20 per cent between the duplicates is therefore not so significant as it might appear at first glance.

⁴ From the few data available on the leachings of the B₂ horizon, it would seem that the quantity filtering through this horizon would not markedly upset the calculations made on the movement of the constituents through the profile.

while. An analysis of the climatological data, however, revealed that the peaks in the Cl curve correspond to rainfall from the geographic supply region of the subtropical air masses which come from the southwest, i.e., from the Atlantic. The water particles carried with these winds bring the Cl and, for that matter, every element found in ocean waters. On the other hand, the rains coming from the northwest carry water particles from the Great Lakes

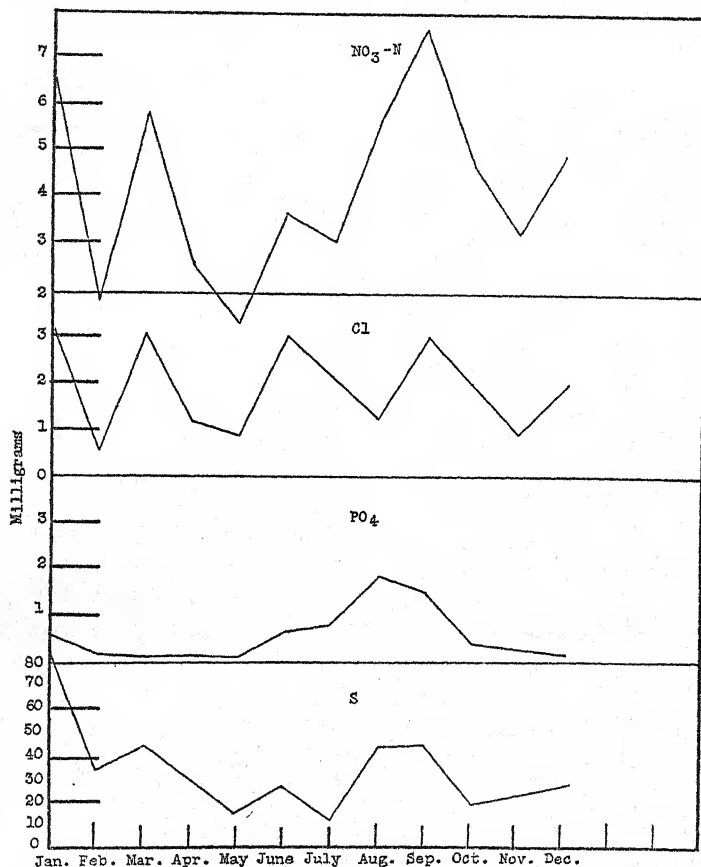


FIG. 1. MOVEMENT OF ANIONS THROUGH THE A₁ HORIZON
Average of 7 years

regions, which contain no chlorine. Thus the data on the dips and peaks may serve as guides to the types of prevailing storms.

Nitrates

No other constituent of the soil has received so much attention as have the nitrates. Through the combined researches of the plant physiologist and the microbiologist, the nitrification reactions and the utilization of nitrates

by plants have been synchronized. It is well known that during early summer large quantities of nitrates are formed, but they are used up by the plants as fast as they are formed.

The regularity of the seasonal nitrate peaks has been shown by the Rothamsted and other experiments, and yet Russell (5, p. 378) says, "Unfortunately it is not possible to form any estimate of the amount of nitrate washed out by rain water." The nitrate-nitrogen curve of figure 1 corroborates the Rothamsted findings that during most of the spring and summer the nitrate content is low. This is due to the rapid utilization of the nitrates by the active growth of the plants during this period. During early fall there is a sudden rise in nitrification, which disappears soon, to reoccur in December and January and again during the spring awakening before plants become active. It seems as if nature prepares nitrogen for the early spring when plants begin to be active.

The remarkable similarity in the peaks and dips of the curves of the $\text{NO}_3\text{-N}$ and Cl makes one wonder whether there is any relation between the movement of these two anions. Does a high Cl content stimulate nitrification or does it prevent the utilization of nitrates? And is it not possible that the other trace elements that come with ocean water, like B , I , Br , also have an influence on the nitrate cycle?

As to the question raised by Russell, an examination of the data in table 1 shows that close to 7 pounds of nitrate nitrogen per acre passes through the A_1 horizon. About one-third of that passes through the A_2 horizon; the rest apparently is used up by the plants. In other words, only about 2.3 pounds (the average of the N from the A_2 leachings) of nitrate nitrogen escapes the rhizosphere after passing a depth of 42 cm. This calculation is correct, provided the aforementioned supposition concerning the vertical movement of water through the A horizon is true. It also shows that under conditions of forest growth very little nitrate nitrogen is lost from the sphere of root distribution. The data on the nitrogen from the B horizon tell us little, since the 0.30 pound of nitrogen found in the leachings does not represent the entire amount that passes through this horizon: no data are available on that which passes through channels.

Sulfur

The data in table 1 show that 28.4 pounds of sulfur goes through the A_1 horizon, a little less than half (the average of the two A_2 funnels) passes through the A_2 horizon, and only a small quantity filters through the B_1 horizon.

The concentration of sulfur in the leachings from the various horizons differs; in the A_2 horizon it is approximately twice that in the A_1 and B_1 , in which the concentrations are about equal. This would indicate that the plants utilize the sulfur from the B_1 horizon, otherwise the concentration would have been higher.

From the curve on the sulfur translocation (fig. 1) it is clear that, in general, the lowest quantities of sulfur appear during the May-July and October-De-

ember periods. The appearance of the largest quantities of sulfur during the December-February period may be explained by the abundance of sulfur in the air from the burning of coal. The slight rise in the curve during the months of August and September may be ascribed to the release of the sulfur compounds from the partial leaf fall which usually takes place during this period. Why the rise does not extend through October when most of the leaves fall is difficult to explain. The decline of sulfur in the leachings during the late spring and summer months is undoubtedly due to the utilization of this element by the plants.

Phosphorus

Phosphorus is one of the nutrient elements generally known for its immobility in soils. Pedologists, however, point out that under natural conditions this element accumulates in the B horizon of the soil zonal type under discussion. The data in table 1 show a definite translocation of this element, but the quantity translocated is very small. Only 0.33 pound of phosphorus passes through the A_1 horizon; about one-fifth of that passes through the A_2 horizon; and, in turn, about one-third of that passing through the A_2 is recovered in the leachings of the B horizon.

The small quantities of soluble phosphorus available in the leachings raise the question of the supply of this nutrient to the forest flora. Because of the low solubility products of the phosphates that occur in the soils under consideration, the concentration is low at any one moment and yet apparently sufficient to supply the demand of the plants. One may, in this case, see the possibility of contact exchange as expounded by Jenny and Overstreet (1).

From the curve on the PO_4 translocation (fig. 1) it is evident that during July, August, and September there is a slight increase in the movement of phosphorus in the A_1 horizon. Why such an increase occurs cannot be explained. Is this period one of low utilization by the forest flora? It is hard to conceive of any period during which the low concentration of phosphorus in the soil solution should be in excess of the demands of the flora. It is possible that the relatively high phosphorus content during this period is due to the contamination of arsenic in the leachings. During these months heavy sprayings with arsenicals against Japanese beetles have been made in the orchard adjacent to the wood where the lysimeters are located.

Silica

The quantity of silica moving through the profile is fairly high, as indicated in table 1. What happens to the 8.77 pounds of SiO_2 that moves through the A_1 horizon after it passes through the A_2 horizon is problematic. Only about 2.81 pounds (the average of the duplicates) is recovered in the leachings of A_2 . Does the rest accumulate in the A_2 horizon? Is it utilized by the forest flora? From the pedologic point of view, the accumulation of SiO_2 in the A_2 horizon is a natural phenomenon of the process of podzolization.

This accumulation, however, is in a large measure attributed to the removal of bases from this horizon. The data of the leachings, however, cannot be disregarded in this connection.

Of the 2.81 pounds per acre that reaches the A_2 horizon, 1.61 is recovered in the leachings of the B_1 . How much drops through the channels of this horizon is not known.

Much of the SiO_2 may remain in the B horizon, $2.81 - 1.61 = 1.20$ pounds. The process of "new formations," i.e., the chance for SiO_3^- to combine with Ca^{++} , Mg^{++} , Fe^{+++} , or Al^{+++} , which takes place in the podzol zone, should be considered.

GENERAL DISCUSSION

In tracing the movement of the anions through the profile one must consider the balance sheet of the intake and outgo of ions whenever possible.

It is difficult to establish the balance for nitrogen, as we cannot evaluate accurately the quantity fixed; we do not know exactly the quantity added by precipitation, although fairly reliable figures seem to indicate that the amount is not less than 5 pounds per acre (4); in the case of forest vegetation, we cannot establish, with any degree of accuracy, the nitrogen tied up by the growing plants; we do not know the run-off losses; and we are not sure that nitrogen, as the gas, is lost through denitrification. It is obvious from the data, however, that not much nitrate nitrogen is lost, assuming that the nitrates which pass beyond the A_2 horizon are lost. About 2.3 pounds of NO_3 nitrogen passed through the A_2 horizon. Since the nitrates make up about 50 to 75 per cent of the nitrogen⁵ passing through the A_2 horizon, as shown previously (4), it is clear that 3 to 4.6 pounds of total nitrogen passes through this horizon. Remembering that some nitrate nitrogen is retained by the B horizon for plant use, we may fairly conclude that not much nitrogen is lost from the soil body by leaching under forest conditions. No data by the method of the funnel type of lysimeter are available on the movement of nitrates in cultivated soils. Undoubtedly more is lost; how much more, is not known.

What has been said of the balance sheet of nitrogen is in a large measure true of the sulfur, except that there is no fixation of this element. The quantity coming down in precipitation depends on the air contamination, as has been pointed out. It is of interest to note that in comparison with the other anions considered, relatively small quantities of sulfur leach out, as indicated by the analyses of the B horizon leachings. This points to a high activity of the roots in this horizon.

The balance sheet of phosphorus is very interesting. There is no outside source to replenish the loss. Under natural conditions one would therefore expect the intake by plants to about balance the outgo. In other words, the

⁵ The rest of the nitrogen disappears in the form of organic nitrogen compounds. A little NH_4 nitrogen passes beyond the B horizon. Positive tests for NH_4 were obtained on the B horizon leachings.

roots pick up enough to satisfy the needs of the flora, and the dead organic matter returns a similar quantity of phosphorus. It is fortunate that this element forms very insoluble compounds. The soil is thereby able to keep up the phosphorus balance, and little or no loss of phosphorus occurs.

The balance sheet of Cl and SiO_2 is not so important from the standpoint of the plant-soil system, and need not be emphasized here.

If we consider the movement of the various anions from one horizon to another, the following picture is obtained for the A horizon: about 42 per cent of the S from the A_1 leaches through the A_2 ; Cl is next with 40 per cent; $\text{NO}_3\text{-N}$, 34 per cent; SiO_2 , 32 per cent; and P, 17 per cent. The B horizon presents the following picture: 59 per cent of the SiO_2 which leaves the A_2 horizon leaches through the B_1 ; 33 per cent of the Cl; 27 per cent of the P; 17 per cent of the S; and 14 per cent of the $\text{NO}_3\text{-N}$.

Though no definite trends may be inferred from the calculations, they do show which of the anions linger more in the A_1 or A_2 horizon; they also show from which horizon the anions are likely to be utilized by plants. The bulk of the nutrients appears to be utilized by the plants in the A_2 horizon. This has a bearing on the root distribution of plants. Thus it is in this horizon that root activity is high. A check-up of a series of forest sections, both natural and planted, in the brown podzolic soils of New Jersey has proved that the A_2 and the upper portion of the B horizon are the regions of greatest root activity. In the true podzols the A_1 horizon has the highest concentration of roots.

The knowledge gained from the data presented has been successfully applied in other fields of soil studies. A specific case is the experimental work in collaboration with the horticultural department on the distribution of roots of apple trees in the soil profile. This work suggested methods of treating shallow-rooting fruit trees of old and, more especially, of new orchard plantings. These findings will be published in due course.

REFERENCES

- (1) JENNY, H., AND OVERSTREET, R. 1939 Cation interchange between plant roots and soil colloids. *Soil Sci.* 47: 257-272.
- (2) JOFFE, J. S. 1932 Lysimeter studies: I. Moisture percolation through the soil profile. *Soil Sci.* 34: 123-143.
- (3) JOFFE, J. S. 1933 Lysimeter studies: II. The movement and translocation of soil constituents in the soil profile. *Soil Sci.* 35: 239-257.
- (4) JOFFE, J. S. 1933 Lysimeter studies: III. The movement and translocation of nitrogen and organic constituents in the profile of a podzolic soil. *Soil Sci.* 35: 401-411.
- (5) RUSSELL, E. J. 1937 Soil Condition and Plant Growth, ed. 7. Longmans, Green, and Co.



THE LAWS OF SOIL COLLOIDAL BEHAVIOR: XXII. THE THERMAL STABILITY OF ACIDOIDS AND BASOIDS

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The amphoteric reactions of a soil, as expressed by an exchange acidity or an exchange alkalinity in salt solutions of various valence combinations (i.e., BaCl_2 , Na_2SO_4), in many instances show no relationship to the $\text{SiO}_2/\text{M}_2\text{O}_3$ ratio in the soil colloidal fraction. This is due, among other things, to the fact that the soil contains more or less humus acidoids which are very active and which neutralize, to a large extent, the mineral basoids of the soil. Thus a brown earth may yield as high an exchange acidity or as low an exchange alkalinity in the A horizon as a podzol in the A_2 horizon, although the $\text{SiO}_2/\text{M}_2\text{O}_3$ ratio may be much higher in the latter. The podzol has lost sesquioxides by eluviation, whereas the brown earth has accumulated more humus acidoids. The acidoid/basoid activity ratio may therefore be about the same in both and give rise to the same reaction.

If it were possible to remove or destroy the humus acidoids without impairing the activity of the mineral acidoids and basoids, the amphoteric reactions of the remaining soil material would be an expression of the acidoid/basoid activity ratio of the mineral fraction. The differences in the amphoteric reactions between a cationically eluviated soil, i.e., a podzol, and an anionically eluviated soil, i.e., a brown or red earth or a laterite, would then be greater, and we would have a simple and practical as well as scientific method for characterizing such soils.

Recalling the work of Kelley, Dore, and Brown (2), who found no loss in exchange capacity after heating mineral colloids to 350°C ., we decided to study the effect of heating.

Table 157 shows the effect of heating for 6 hours at various temperatures on certain soil characteristics. The soil material is a B_2 -horizon sample of the Håggbygget podzol (1).

The study included the loss on ignition, the amount of KMnO_4 required to oxidize the soil, the cation-exchange capacity by the Ca-acetate method, the acid-oxalate-soluble sesquioxides according to Tamm (6), and the pH of the soil in water and in 0.01 N Na_2SO_4 and BaCl_2 . The three pH deter-

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TABLE 157

Changes in a soil material (B₂ horizon, Haggbygget podzol) after 6 hours' heating at various temperatures

TEMPERATURE	LOSS IN WEIGHT	KMnO ₄ -VALUE*	EXCHANGE CAPACITY	ACID-OXALATE-SOLUBLE			pH IN		
				SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	Water	0.01 N Na ₂ SO ₄	0.01 N BaCl ₂
°C.	per cent	cc./gm.	m.e./100 gm.	per cent†	per cent†	per cent†			
—	46.0	13.3	0.26	1.39	2.77	4.41	4.65	4.01
100	46.2	13.3	0.17	4.27	4.67	4.03
150	1.29	37.2	8.7	0.15	1.11	2.90	4.16	4.33	4.06
200	5.37	27.1	1.53	0.17	1.33	2.66	4.30	4.49	4.21
250	6.16	11.5	0.90	0.15	1.17	2.75	4.46	5.27	4.41
300	6.47	7.5	1.01	0.15	0.76	3.07	4.58	5.68	4.65
350	6.67	6.2	1.27	0.15	0.75	2.66	4.55	5.82	4.70
400	6.98	4.2	1.47	0.17	1.05	2.07	4.75	5.85	4.65
460	7.07	1.5	0.91	0.17	0.85	1.49	5.64	6.03	4.73
520	7.17	0.5	0.88	0.16	0.61	1.32	5.84	6.03	4.85
570	7.52	0.6	0.63	0.14	0.25	0.98	5.63	5.82	4.67
700	7.77	0.5	1.12	0.12	0.01	0.34	5.49	5.52	4.72
880	7.78	0.2	0.10	0.04	0.02	0.04	6.82	6.52	6.07

* 0.1 N KMnO₄ required to oxidize the soil.

† On the basis of ignited material.

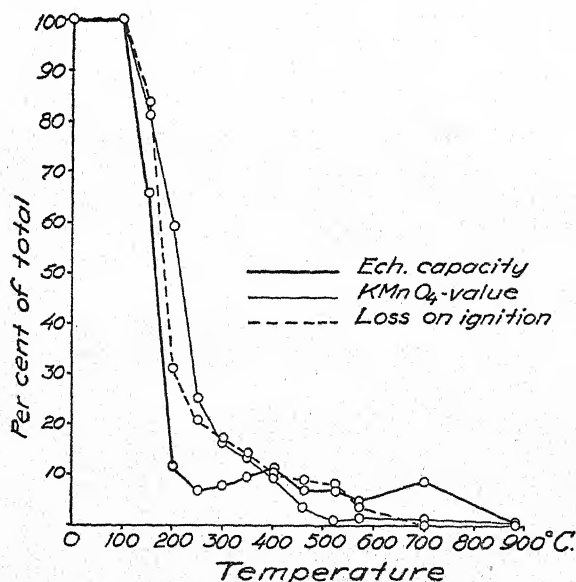


FIG. 102. PERCENTAGE CHANGE IN THE EXCHANGE CAPACITY, KMnO₄-VALUE, AND LOSS ON IGNITION OF A SOIL AFTER HEATING TO VARIOUS TEMPERATURES

minations were always made at the same time and with the greatest possible care with the quinhydrone electrode.

The exchange capacity, which amounts to 13.3 m.e. per 100 gm. original soil is reduced to 8.7 m.e. after heating to 150°C. and to about 1 m.e. after heating to 250° or more. It will be noted that the power to bind cations falls off more rapidly than the loss on ignition and the KMnO_4 -values (fig.

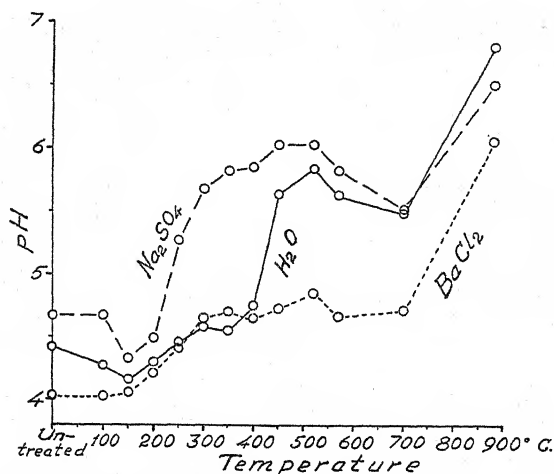


FIG. 103. THE pH OF THE HEATED SOIL IN WATER AND IN 0.01 N Na_2SO_4 AND BaCl_2 SOLUTIONS

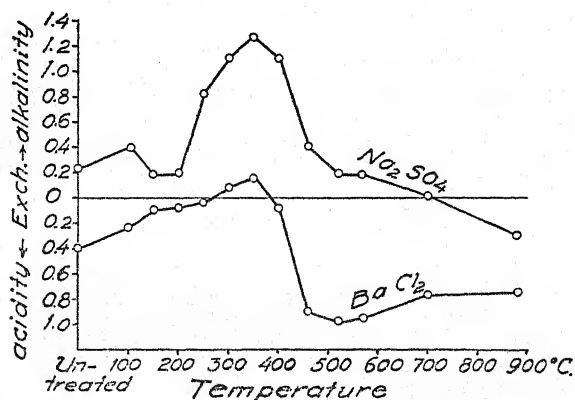


FIG. 104. EXCHANGE REACTIONS OF THE HEATED SOIL IN 0.01 N Na_2SO_4 AND BaCl_2 SOLUTIONS

102). This may be taken to indicate that the acidoid groups ($-\text{COOH}$) are more readily oxidized than the rest of the organic complex.

The mineral acidoids seem to survive a temperature of 700°C. At 880° even these acidoids lose the power of base exchange, evidently because of dehydration. In this connection, the small amount of SiO_2 dissolved in the sample heated to 880°C. should be noted.

Figure 103 shows graphically how the heating has affected the pH, and fig-

ure 104 gives the differences between the pH in water and the pH in the salt solutions, expressed as exchange alkalinity when the pH is higher in the salt solution and as exchange acidity when it is lower than the pH in water.

The following facts will be noted:

The untreated soil yields a fairly small exchange alkalinity (= 0.24 pH unit) in the Na_2SO_4 solution and a moderate exchange acidity (= 0.40 pH unit) in the BaCl_2 solution.

Heating to 150° causes a loss in the exchange alkalinity (which falls to 0.17 pH unit), as if a part of the basoids were particularly sensitive to heating. (We shall later show that nascent basoids are very sensitive.)

The sample heated to 200° shows no significant increase in exchange alkalinity, although the power to bind base (fig. 102) has been reduced to a very low value. This would seem to indicate that the free humus acidoid residue is destroyed before the acidoid in combination with the basoid is destroyed.

The samples heated to 250, 300, 350, and 400° show an increase in all the pH values and yield exchange alkalinities in the sulfate solution amounting to 0.81, 1.10, 1.27, and 1.10 pH units respectively. Even in the chloride solution the heated soil yields a small exchange alkalinity in the case of two samples. This must all be ascribed to a destruction of the humus acidoids in combination with the sesquioxide basoids, thus setting the latter free to combine with the anions of the salt.

At a temperature of 460° there is a sharp decline in the exchange alkalinity due, it may be assumed, to the inactivation of the basoids. There is at the same time a sharp increase in the pH of the soil in water. This increase is difficult to explain in view of the fact that the pH in the BaCl_2 solution remains low, indicating that the mineral acidoids are still active (the formation of strong base by calcination of some silicate might be suggested as an explanation).

The exchange alkalinity in the sulfate solution falls to a very low value between 460 and 700° , whereas the exchange acidity in the chloride solution remains high even in the sample heated to 880° . This, together with the fact that the latter sample yields an exchange acidity in the sulfate solution, indicates a great heat resistance of the mineral acidoids. The fact that all three of the pH values of this sample are very high (above 6) shows, however, that the acidoids are here extensively inactivated. The high exchange acidity of this sample in the chloride solution (0.75 pH unit) is no indication of a high acidoid content. In the absence of active basoids, very little acidoid is required to produce a considerable exchange acidity at a pH so near the neutral point. A sand may yield as high an exchange acidity as that of a clay.

If we plot the sum of the acid-oxalate-soluble sesquioxides against the temperature, we get the curve in figure 105. It is interesting to note that the solubility of the sesquioxides is related to their activity as basoids. Thus between a temperature of 400 and 500 there is a rapid decline in both of these properties. This is, of course, to be expected, because the solubility in the oxalate solution and the exchange alkalinity in the sulfate solution are both the result of a union between the anions of the solutions and the Al and Fe of the soil. The dehydrated sesquioxides are inactive toward both reagents.

APPLICATION TO SOIL PROFILES

The podzol

This method of studying the amphoteric reactions of the mineral soil complex by a preliminary destruction of the soil humates was applied to soil

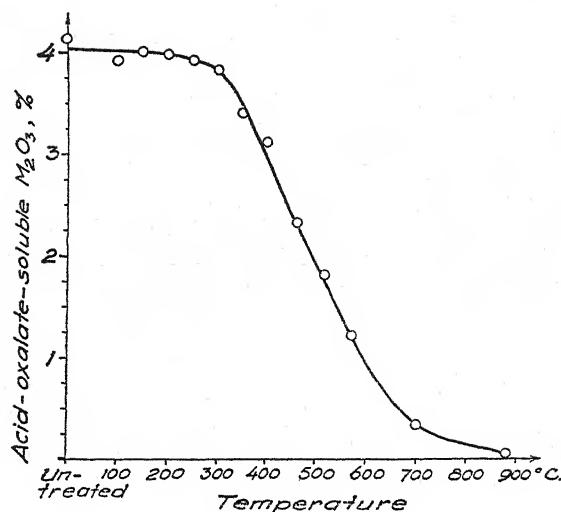


FIG. 105. ACID-OXALATE-SOLUBLE SESQUIOXIDES IN THE HEATED SOIL

TABLE 158

Some properties of the samples of the Häggbygget podzol and the Dala brown earth profiles before and after heating for 6 hours at 275°C.

HORIZON	LOSS ON IGNITION	EXCHANGE CAPACITY	ACID-OXALATE-SOLUBLE			pH IN		
			SiO ₂	Al ₂ O ₃	Fe ₂ O ₃	Water	0.01 N Na ₂ SO ₄	0.01 N BaCl ₂
	per cent	m.e./100 gm.	per cent*	per cent*	per cent*			
Untreated podzol								
A ₁	55.2	3.21	3.05	2.81
A ₂	1.47	2.4	0.14	0.10	0.05	4.04	3.85	3.29
B ₁	9.02	24.8	0.40	0.98	2.36	3.97	3.97	3.46
B ₂	7.77	13.0	0.26	1.39	2.77	4.41	4.65	4.01
B ₃	3.46	4.6	0.37	1.07	0.53	4.68	4.91	4.35
Heated podzol								
A ₁	3.6	6.21	6.25	6.06
A ₂	0.58	1.9	0.14	0.06	0.07	4.06	4.01	3.50
B ₁	1.70	3.1	0.18	0.91	1.82	4.44	5.11	4.33
B ₂	1.50	0.9	0.15	0.98	2.91	4.52	5.62	4.52
B ₃	1.35	0.8	0.33	0.93	0.50	4.89	5.75	4.75
Untreated brown earth								
A	7.56	9.3	0.09	0.42	1.00	4.74	4.65	4.12
B ₁	3.40	4.4	0.14	0.62	1.05	4.94	4.92	4.27
B ₂	1.51	4.0	0.17	0.67	1.56	4.87	4.98	4.21
Heated brown earth								
A	2.08	3.5	0.12	0.38	0.98	5.01	5.07	4.56
B ₁	1.40	2.5	0.14	0.53	1.25	5.18	5.32	4.58
B ₂	1.31	2.3	0.16	0.41	1.92	5.06	5.31	4.47

* On the basis of ignited material.

profiles by heating the samples for 6 hours to a temperature of 275°C. The Häggbygget podzol and a brown earth from an old pasture on a stream terrace in Dala near Båstad in southern Sweden were studied in some detail. Table 158 gives the analytical data.

Figure 106 shows graphically how the base-exchange capacity is affected by the heating of the podzol samples. In the A_1 , A_2 , and B_1 horizons this capacity is appreciable even after the heating and must be ascribed to the silicate acidoid. We note, however, that there is no relationship between this and the acid-oxalate-soluble SiO_2 . But acid-oxalate is no solvent for silica and is in this connection of no importance. The major part of the silicate acidoid occurs in the "micropedosphere" on the individual soil particles and remains there after the oxalate treatment, which generally leads to an increase in the base-exchange capacity (3).

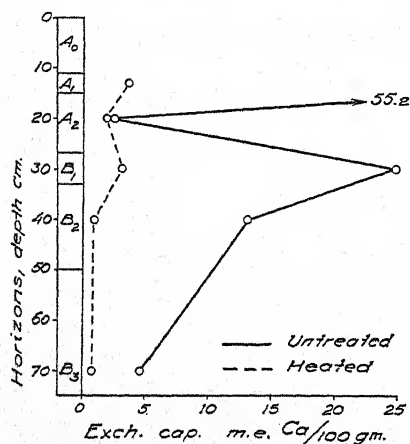


FIG. 106. EXCHANGE CAPACITY OF THE PODZOL SAMPLES BEFORE AND AFTER THE HEATING

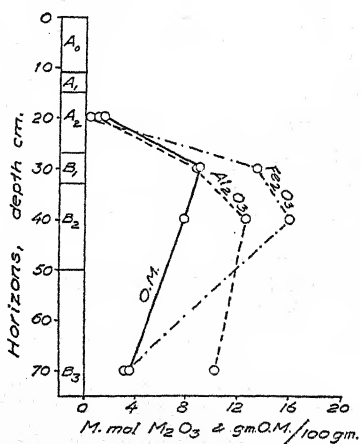


FIG. 107. ACID-OXALATE-SOLUBLE SESQUIOXIDES AND LOSS ON IGNITION (=O.M.) IN THE PODZOL PROFILE

Figure 107 shows the loss on ignition (approximately equal to organic matter = O.M.) expressed in grams, and the sesquioxides expressed in millimols, per 100 gm. dry soil in the podzol profile (these units are convenient and make possible a direct comparison with Mattson's earlier work on the amphoteric humates, cf. fig. 108). In accordance with the theory of isoelectric weathering, the upper B horizons are richer in humus and Fe, whereas the lower B_3 is richer in Al: the ferric humates have a lower isoelectric point than the Al-humates of corresponding composition and must, therefore, be the first to precipitate in a profile the pH of which increases with depth.

If we plot the ratios of grams humus to millimols sesquioxides against the pH (= approximate ultimate pH = approximate isoelectric point) in the podzol profile together with the same ratio against the isoelectric pH of Mattson's (1) isoelectrically precipitated humates, we get the results shown in

figure 108. The podzol curve corresponds more nearly to the Fe-humate than to the Al-humate curve. This is obviously in part due to the fact that the podzol complex contains some silicate acidoid in addition to the humus acidoid.

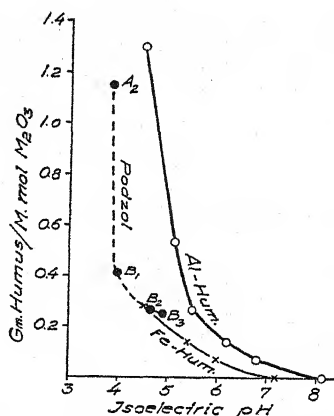


FIG. 108. THE COMPOSITION OF THE PODZOL GEL COMPLEX AGAINST THE PROFILE pH, COMPARED TO THE COMPOSITION OF THE ISOELECTRICALLY PRECIPITATED ALUMINUM AND FERRIC "HUMATES" AGAINST THEIR ISOELECTRIC pH

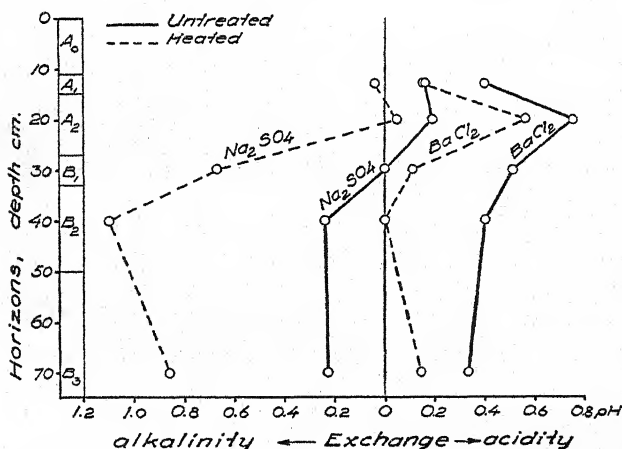


FIG. 109. EXCHANGE REACTIONS OF THE PODZOL SAMPLES IN 0.01 *N* SOLUTIONS OF Na_2SO_4 AND BaCl_2 BEFORE AND AFTER THE HEATING

Figure 109 shows the exchange reactions in the two salt solutions before and after the heating. The effect of the heating is greatest in B_2 , which shows the greatest basoid activity. In comparing figures 107 and 109 it must be remembered that alumina has a higher isoelectric point than ferric hydroxide and that at a given pH, i.e., that of the soil, the former is, therefore, more

active. Thus, for example, although the sum of the sesquioxides in B_2 is more than double the sum in B_3 , the basoid activity, as expressed by the exchange alkalinity of the heated samples in Na_2SO_4 solution (fig. 109), is not twice as great in B_2 because the B_3 , although much poorer in Fe, contains nearly as much Al.

The exchange reactions in a podzol profile after the humus acidoids have been destroyed by heating the soil are apparently very effective in establishing clearly the differentiation brought about by the cationic solvation and precipitation. The chemical analysis measures the mass, whereas the exchange reaction measures the activity, and the latter is more important in characterizing the soil.

In applying the method of exchange-reaction determination, the following facts must be borne in mind: The exchange reaction of a soil material in a given salt solution (as compared to the reaction in water) is determined by (5) the saturation of the soil with acid or base, the concentration of the salt solution, the activity ratio of acidoids to basoids, and the concentration of the colloid.

As an example let us consider the fluctuations in the exchange alkalinity in the 0.01 N Na_2SO_4 solution.

Differences in exchange alkalinity may be due to differences in base saturation: a soil saturated with a certain amount of base does not exchange its OH ions for the anions of the salt and does not, therefore, yield any exchange alkalinity. Differences in exchange alkalinity may also be due to differences in the acidoid/basoid ratios as is always the case in the podzol profile. But since the maximum in exchange alkalinity occurs at a lower concentration of Na_2SO_4 the lower the proportion of basoids to acidoids, the fluctuations in exchange alkalinity will be greater when determined by the use of one single concentration of salt (i.e., 0.01 N) instead of a concentration which produces a maximum in exchange alkalinity in each sample. Thus, where the proportion of basoids is very low, a 0.01 N solution of Na_2SO_4 may produce an exchange acidity, whereas a more dilute solution will yield an exchange alkalinity. Differences in exchange alkalinity may, finally, be due to differences in colloid content, but here again it must be borne in mind that the maximum in exchange alkalinity occurs at a lower concentration of Na_2SO_4 the lower the colloid content of the soil.

From what has been said it is clear that the results obtained in a study of the exchange reactions in a soil profile are strictly comparable only if all the samples are virtually completely unsaturated and if the pH determinations are made in that concentration of the salt solutions which produces a maximum in valence effect as expressed by an exchange alkalinity or exchange acidity. Finding the maximum effect would, however, require a much greater number of pH determinations than were made during the present work. It should finally be pointed out that while we are employing 0.01 N salt solutions the final equilibrium concentration (and this is what counts)

will be different and will vary from case to case depending upon the amount of exchange which, at the equi-ionic point, may be considerable because of the formation of H_2O .

The brown earth

Turning our attention to the brown earth, we note from the analysis that the acid-oxalate-soluble sesquioxides show a fairly uniform increase with depth (fig. 110). This is probably due to other causes (parent material, mechanical washing, etc.) than podzolization, for which the pH is too high.

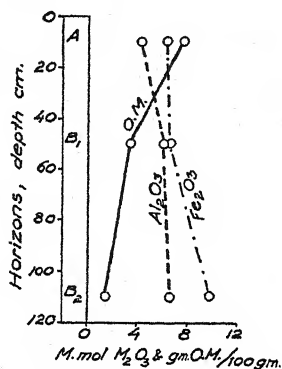


FIG. 110. ACID-OXALATE-SOLUBLE SESQUIOXIDES AND LOSS ON IGNITION (=O.M.) IN THE BROWN EARTH PROFILE

TABLE 159

The pH of the electrodialyzed samples of the Dala brown earth before and after the heating

HORIZON	pH IN WATER		pH IN 0.01 N Na_2SO_4		pH IN 0.01 N BaCl_2	
	Before	After	Before	After	Before	After
A	3.90	4.75	4.13	4.95	3.67	4.31
B ₁	4.51	5.14	4.71	5.24	4.11	4.47
B ₂	4.49	5.16	5.00	5.47	4.20	4.47

This increase in sesquioxides with depth is reflected in the exchange reactions of both the untreated and the heated samples in the Na_2SO_4 solution (fig. 111). The exchange acidity decreases and the exchange alkalinity increases in this solution from A to B₂. The deviation from this trend in the BaCl_2 solution is probably due to the small amounts of SO_4 present in the exchangeable form in the B horizons (CaCl_2 ought to have been used).

Activation through electro dialysis

The Dala brown earth is almost base unsaturated, but in order to remove the small amounts of displaceable ions the samples were electro dialyzed.

The pH values of the electrodialyzed samples are given in table 159, and the exchange reactions are shown graphically in figure 112.

The results were unexpected. The increase in exchange alkalinity and the decrease in exchange acidity which were caused by the base desaturation are normal phenomena, but the fact that the heating of the electrodialyzed soil desactivates rather than activates the basoids was new to us. It teaches us that electrodialysis of the soil activates the sesquioxides and that the nascent basoids are very sensitive to heating and are more easily desactivated than the naturally occurring species of basoids (cf. fig. 111 and 112).

Electrodialysis is a process which brings about the same changes in the soil as does either laterization or podzolization, depending upon the pH (4). If the pH is low, the sesquioxides are mobilized, migrate toward the cathode, and become enriched in the soil (if this forms a stationary layer) next to the

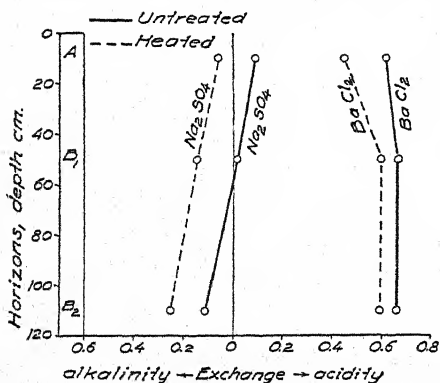


FIG. 111. EXCHANGE REACTIONS OF THE BROWN EARTH SAMPLES IN 0.01 *N* SOLUTIONS OF Na_2SO_4 AND BaCl_2 BEFORE AND AFTER THE HEATING

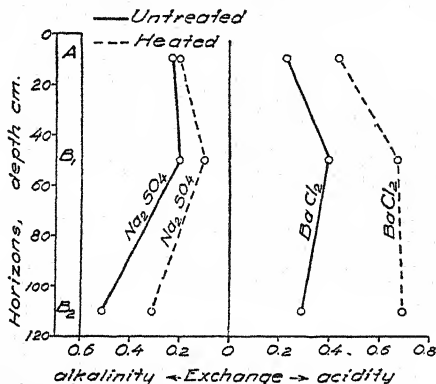


FIG. 112. THE SAME AS FIGURE 111, BUT THE SAMPLES WERE FIRST ELECTRODIALYZED. NOTE THE REVERSED EFFECT OF THE HEATING

cathode membrane, thus forming a layer which corresponds to the B horizon in the podzol profile. The basoid activation should therefore be greatest in this layer.

To test this, we electro-dialyzed the Dala B₂ sample again for several days in the so-called Mattson cell. We then divided the contents of the 2.5-cm. soil compartment into the half adjacent to the cathode membrane and the half adjacent to the anode membrane. The pH values of the two samples before and after heating to 275°C. for 6 hours are given in table 160.

The results show that the pH is higher and the basoid activity is greater in the half adjacent to the cathode than in the half adjacent to the anode membrane; the heating increases the pH but desactivates the basoids, despite the destruction of the humus acidoids; and this desactivation is greater in the cathode sample, which contains the greatest amount of nascent basoids.

TABLE 160

The pH and exchange reactions before and after the heating of an electrodyalized Dala B₂ sample;
A adjacent to the anode and C adjacent to the cathode membrane

SAMPLE	pH IN			EXCHANGE	
	Water, <i>w</i>	0.01 <i>N</i> Na ₂ SO ₄ , <i>s'</i>	0.01 <i>N</i> BaCl ₂ , <i>s''</i>	Alkalinity, <i>w - s'</i>	Acidity, <i>w - s''</i>
<i>Before heating</i>					
A	4.25	4.67	3.90	0.42	0.35
C	4.60	5.21	4.30	0.61	0.30
<i>After heating to 275°C.</i>					
A	5.02	5.33	4.49	0.31	0.53
C	5.18	5.56	4.65	0.38	0.53

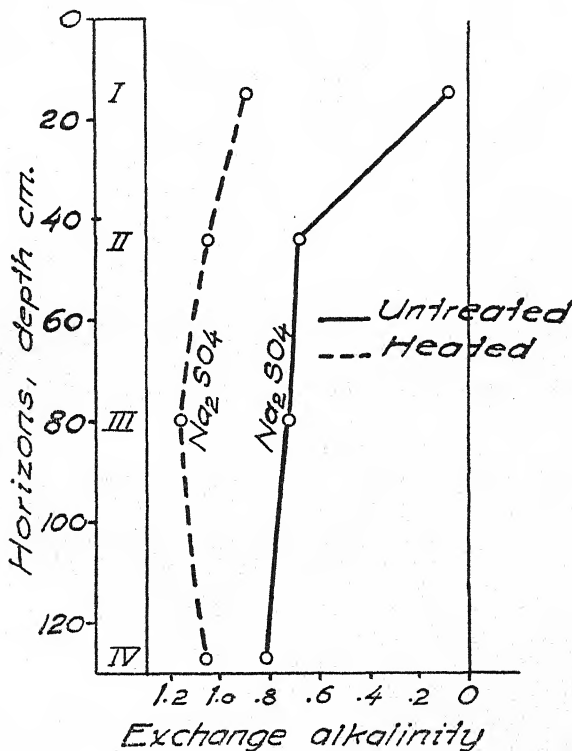


FIG. 113. EXCHANGE ALKALINITY OF FOUR SAMPLES OF THE LAS MESAS LATERITE IN *N* Na₂SO₄ SOLUTION BEFORE AND AFTER THE HEATING

The observed differences would undoubtedly be greater in a heavy soil. Thus in the electrodyalized Sharkey clay soil, Mattson (4) found a pH of 3.4 next to the anode membrane and pH 6.0 next to the cathode membrane.

The experiment emphasizes the fact that electrodialysis of the soil brings about fundamental changes in addition to the removal of the exchangeable ions. But the experiment also suggests important applications of the principles of exchange activity to a study of the state and of the transition points of the hydrous oxides and of acidoids and basoids in general.

The laterite

In the preceding paper (5) we included the Las Mesas laterite in our investigation. No additional work has been done on this soil other than a determination of the pH by the glass electrode of the heated samples in N Na_2SO_4 solution. A normal solution was used because, in strongly basoid soils, a concentrated solution produces the greatest effect. The results are shown in figure 113.

The heating has increased the exchange alkalinity of every sample, thus indicating the presence of humus acidoids at considerable depths of the profile. The increase is greatest, however, in the surface sample. The exchange alkalinity of this sample, nevertheless, stays below that of the other samples. This might be an expression of a lower basoid activity in the surface horizon as indicated by the presence of concretions.

SUMMARY

The thermal stability of soil acidoids and basoids seems to place them in the following order: free humus acidoids < sesquioxide-bound humus acidoids < sesquioxide-basoids < silicic acidoids.

As a result of destruction of the humus acidoids by heating for 6 hours to $275^\circ\text{C}.$, the exchange reactions of the soil become an expression of the mineral acidoid/basoid activity ratio and serve to characterize the soil type.

REFERENCES

- (1) BORATYŃSKI, K., AND MATTSON, S. 1938 The electro-chemistry of soil formation: III. The humate complex. *Ann. Agr. Col. Sweden* 7: 65-119.
- (2) KELLEY, W. P., DORE, W. H., AND BROWN, S. M. 1931 The nature of the base-exchange material of bentonite, soils, and zeolites, as revealed by chemical investigation and X-ray analysis. *Soil Sci.* 31: 25-55.
- (3) LUNDBLAD, K. 1934 Studies on podzols and brown forest soils: I. *Soil Sci.* 37: 137-155.
- (4) MATTSON, S. 1933 The laws of soil colloidal behavior: XI. Electrodialysis in relation to soil processes. *Soil Sci.* 36: 149-163.
- (5) MATTSON, S., AND WIKLANDER, L. 1940 The laws of soil colloidal behavior: XXI. The amphoteric points, the pH, and the Donnan equilibrium. *Soil Sci.* 49: 109-153.
- (6) TAMM, O. 1932 Über die Oxalatmethode in der chemischen Bodenanalyse. *Meddel. Statens Skogsförsöksanst. [Sweden]* 27: 1-20.

BOOKS

An Outline of British Crop Husbandry. By H. G. SANDERS. Cambridge University Press, 1939. Pp. 348, illus. 6. Price, \$4.50.

A book on the agriculture of Great Britain, giving consideration to rotating, manuring, seeding, cultivating, and harvesting the crops.

Botany. Third Edition. By WILLIAM J. ROBBINS AND HAROLD W. RICKETT. D. Van Nostrand Company, Inc., New York, 1939. Pp. 658, illus. 440. Price, \$3.75.

A textbook which deals with the living processes in plants in relation to the different botanical groups. The material is presented in a very readable manner. A helpful list of questions covering the topics discussed is appended.

Economics of Food Production. By B. S. ROWNTREE AND VISCOUNT ASTOR. Longmans, Green and Co., New York, 1940. Pp. 251. Price, \$6.

A comparative study of agricultural conditions in Central Europe and those in the British Isles. The means by which production is stimulated and controlled and the restrictions which are placed on imports and exports are discussed. It is pointed out that the simplest means of controlling production is to allow prices to fall and to let each nation produce those products to which its soil, climate, and location are best adapted. But it has become so dangerous for one country alone to be wise that all nations prefer foolishness.

Elements of Farm Management. Revised Edition. By JOHN A. HOPKINS. Prentice-Hall, Inc., New York, 1940. Pp. 489, illus. 89. Price, \$2.20.

The fundamental principles of economic crop and livestock production. Particular attention is directed to the types of farming followed in the north central states.

Field Plot Technique. By WARREN H. LEONARD AND ANDREW G. CLARK. Burgess Publishing Company, Minneapolis, Minn., 1939. Pp. 271. Price, \$3.25.

A manual on field plot technique, in which consideration is given to the problems involved in accurate experimentation and to the analysis of the data obtained. This should be very useful to all station workers engaged in field experimentation. The book is mimeographed.

Gardening Without Soil. By A. H. PHILLIPS. Chemical Publishing Co., Inc., New York, 1940. Pp. 137, illus. 8. Price, \$2.

A concise discussion of methods employed in growing plants in culture solutions, with and without the supplemental use of gravel and similar soil substitutes.

Land Drainage and Reclamation. Second Edition. By QUINCY CLAUDE AYRES AND DANIELS SCOATES. McGraw-Hill Book Company, Inc., New York, 1939. Pp. 496, illus. 316. Price, \$4.

The whole field of land drainage is covered in this book, which takes cognizance, likewise, of the larger problem of water control by means of dams and terraces. Intended primarily for college students, the book will be very useful also to those who are engaged with problems of soil conservation and land utilization.

Land Economics. By RICHARD T. ELY and GEORGE S. WEHRWEIN. The MacMillan Co., New York, 1940. Pp. 512. Price, \$4.

This book deals with the land resources of the United States and the uses to which they are being put. These uses involve not only agriculture, forestry, and grazing, but also the expansion of industry and of urban populations and the exploitation of water and mineral resources. The authors are primarily concerned with the economic and social problems involved in land conservation.

Physical Meteorology. By J. G. ALBRIGHT. Prentice-Hall, Inc., New York, 1939. Pp. 393, illus. 246. Price, \$5.35.

Those interested in the physical principles of meteorology will welcome this book. It gives a more complete presentation of the subject than do most introductory texts. Agronomists will find this precise syllabus very useful.

Plant Biochemistry. Reprint. By WILLIAM EDWARD TOTTINGHAM. Burgess Publishing Co., Minneapolis, Minn., 1939. Pp. 249, illus. 97. Price, \$3.25.

Extensive annual revision of classroom notes over a period of 25 years has resulted in this excellent digest of the literature dealing with the chemical aspects of plant physiology. The book is in mimeoprint form.

Soil Conservation. By HUGH HAMMOND BENNETT. McGraw-Hill Book Company, Inc., New York, 1939. Pp. 993, illus. 358. Price, \$6.

A comprehensive review of the problems involved in soil conservation in the United States, with particular regard to the seriousness of erosion and to methods of erosion control. The book contains a wealth of material which has been brought together for the first time from various sources. Every worker in soil science will want a copy of this book for ready reference.

Soilless Culture Simplified. By ALEX LAURIE. McGraw-Hill Book Company, Inc., New York, 1940. Pp. 201, illus. 22. Price, \$2.50.

The facts about growing plants without soil are presented in such a manner as to overcome the many false notions that are now current on this subject. The book is intended primarily for the amateur but contains practical suggestions of considerable interest to the professional as well.

Soilless Gardening. By WILLIAM F. GERICKE. Prentice-Hall, Inc., New York, 1940. Pp. 285, illus. 60. Price, \$2.75.

The principles and practice of hydroponics as applied to the production of a considerable variety of plants. The author, one of the pioneers in this field, has brought together much useful information bearing on this interesting subject.

The Meteorological Glossary. Third Edition (first American edition). The Chemical Publishing Co., Inc., New York, 1940. Pp. 251. Price, \$3.

The Meteorological Office and the Air Ministry in London present an extremely useful collection of definitions of terms, the meanings of which are necessary for a full understanding of weather maps and forecasts. The book also covers a broad variety of items in related fields, and deals with such practical problems as ice formation on airplanes and the protection of crops against frost. Equivalents of the most important terms are given in nine languages.

Vegetable Crops. Third Edition. By HOMER C. THOMPSON. McGraw-Hill Book Co., Inc., New York, 1939. Pp. 578, illus. 68. Price, \$5.

A comprehensive consideration of the problems involved in the production of vegetable crops, by a man who has had many years of experience in this field. Everyone interested in vegetable growing will want a copy of this book.

THE EDITORS

STUDIES ON ORGANIC PHOSPHORUS COMPOUNDS IN SOIL; ISOLATION OF INOSITOL¹

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Although certain organic phosphorus compounds have been shown to exist in soil, the chemical nature of the major part of the organic phosphorus remains obscure. At various times the organic phosphorus was thought to consist largely of nucleic acid. Shorey (8) isolated a "nucleic-acid fraction" yielding purine and pyrimidine derivatives. More recently, Wrenshall and McKibbin (11) separated a "nucleic-acid fraction" containing 65 per cent of the total organic phosphorus. Previously, Auten (3) had expressed the belief that nucleic-acid compounds represent only a very small fraction of the total organic phosphorus in soil, since the amounts of nitrogen bases which could be isolated were very small. This view was substantiated in a later report by Wrenshall, Dyer, and Smith (10), who found that pyrimidine nucleotides as well as nucleic acid were readily broken down within a few days when incubated in sand cultures. In discussing possible organic phosphorus compounds in soils, Schreiner (7a) mentions phytin and reports that though this has not yet been isolated from soils, some evidence has been obtained in his laboratory of the presence of inosite in soils. The most recent attempt to explain the nature of soil organic phosphorus has been in the direction of phytin. Dyer, Wrenshall, and Smith (5) now propose ferric phytate as the agent responsible for at least part of the bound phosphorus. Their claim to the isolation of phytin from soil is based on phosphorus-iron ratios and resistance to enzyme action.

For the last 2 years an investigation of organic phosphorus in Hawaiian agricultural soils has been in progress. The primary object of this work was to determine, if possible, the chemical nature of the bulk of soil organic phosphorus which so far has escaped identification and to confirm previous findings and views regarding soil organic-phosphorus compounds. In the course of this investigation inositol was isolated for the first time from organic-phosphorus-bearing soil material in a pure crystalline form. Some of the evidence obtained suggests that this compound is present in soil as inositol monophosphate.

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² The writer wishes to acknowledge her indebtedness to L. A. Dean, who suggested and directed this investigation.

EXPERIMENTAL

Three Hawaiian agricultural soils were selected for this study: the first, from a sugar cane field, Honokaa, Hawaii; the second, from a coffee orchard, Kona Branch Station, Hawaii; and the third, from a sudan grass plot, University Farm, Honolulu, Oahu. The percentages of organic matter found in these soils are as follows: Honokaa, 19.6; Kona, 13.9; University Farm, 4.4.

Extraction of the organic phosphorus from soil

The finely ground, air-dry soil was shaken with 10 to 15 volumes of normal hydrochloric acid in an end-over-end shaker for 1 hour. This acid treatment removes such easily soluble bases as calcium and magnesium, the presence of which inhibits the solubility of organic matter. The soil residue was filtered off with suction, washed with dilute hydrochloric acid, and then washed with water until the final wash water was nearly chloride-free. The acid-treated soil was then dispersed in 20 volumes of 0.5 *N* sodium hydroxide and allowed to stand overnight. The dark supernatant alkaline extract was siphoned off

TABLE 1

Amounts of phosphorus in the sodium-hydroxide extract, in α -humus, and in the acid filtrate obtained from 100 gm. of soil

SOIL	SODIUM HYDROXIDE EXTRACT TOTAL P	α -HUMUS TOTAL P	ACID FILTRATE		
			Total P	Inorganic P	Organic P
	mgm.	mgm.	mgm.	mgm.	mgm.
Honokaa.....	164	12	152	55	97
Kona.....	196	8	188	62	126
University Farm.....	423	2	421	385	36

and treated with concentrated hydrochloric acid until the solution was strongly acid (pH 2). The resulting dark precipitate (α -humus) was allowed to settle. The supernatant solution was drawn off and filtered through a pad of paper pulp in a Büchner funnel. The remaining precipitate was then transferred to the funnel and washed with dilute hydrochloric acid and finally with water.

Table 1 shows the total amount of phosphorus extracted by sodium hydroxide and its distribution into acid-insoluble (α -humus) and acid-soluble phosphorus. The amount of acid-soluble organic phosphorus was obtained by subtracting inorganic from total phosphorus in the acid filtrate.

These data show that nearly all the phosphorus extracted by 0.5 *N* sodium hydroxide is soluble at pH 2. In the Honokaa and Kona soils, the acid-soluble organic phosphorus amounted to more than 50 per cent of the total phosphorus extracted by alkali, whereas the alkali-soluble phosphorus of the University Farm soil was chiefly inorganic.

Separation of an organic phosphorus fraction—the "alcohol precipitate"

Since about 90 per cent of the organic phosphorus was found in the acid filtrate, the phosphorus in α -humus was disregarded, and the acid-soluble organic phosphorus was concentrated in the following manner. The acid

filtrate was adjusted to pH 4.2 with dilute sodium hydroxide. By this treatment a precipitate of β -humus containing organic phosphorus was separated from large amounts of inorganic material such as sodium chloride, the latter remaining in solution. After the precipitate settled, the supernatant liquid was drawn off and the precipitate transferred to a Büchner funnel, where it was washed repeatedly with distilled water. An acid filtrate containing 111 mgm. of organic phosphorus obtained from Honokaa soil yielded β -humus containing 96 mgm. of organic phosphorus.

The washed β -humus was then suspended in water and sufficient concentrated hydrochloric acid added to bring the final concentration to 5 per cent by volume (5 cc. of 12 *N* HCl per 100 cc. of solution). After vigorous stirring to break up the lumps, the insoluble material was centrifuged off and the clear acid solution poured slowly with constant stirring into 5 volumes of 95 per cent ethanol. The flocculent precipitate was allowed to settle, the supernatant alcohol siphoned off, and the precipitate transferred to a Büchner funnel. After repeated washings with alcohol, the product was redissolved in 5 per cent hydrochloric acid, filtered, and reprecipitated by pouring the solution into 5 volumes of 95 per cent ethanol. After three such precipitations, no further increase in phosphorus content could be effected by this means. The purified material was dried at 60°C. for several days and then ground to a brown-colored powder. This product contained considerable inorganic matter (21 per cent ash), and was similar to organic phosphorus preparations isolated by other workers (8, 11). In this paper the term "alcohol product" will be used to designate this fraction.

The organic phosphorus contained in the "alcohol product" usually amounted to about 50 per cent of the total organic phosphorus extracted by sodium hydroxide. For example, an alkaline extract from Honokaa soil containing 88.7 mgm. of organic phosphorus yielded an "alcohol product" containing 46.1 mgm. of organic phosphorus. The organic-phosphorus contents of the "alcohol products" obtained from the three soils studied are as follows: Honokaa, 5.8 per cent; Kona, 4.4; University Farm, 4.1.

Previous workers (8, 11) have demonstrated the presence of purine and pyrimidine bases among the hydrolysis products of organic phosphorus material similar to that described above. In order to investigate the presence of these nitrogen bases in the "alcohol product," portions of the material were hydrolyzed for the isolation of pyrimidines and purines by methods outlined by Levene and Bass (6). To isolate purine bases the "alcohol product" was hydrolyzed with 2 per cent sulfuric acid for 2 hours at 125°C. Subsequent addition of silver oxide precipitated a small quantity of silver salts, which were decomposed with hydrogen sulfide. Evaporation of the filtrate yielded a small amount of amorphous material which failed to form a picrate, thus showing the absence of adenine. Another portion of the "alcohol product" was hydrolyzed with 25 per cent sulfuric acid at 175°C. for 2 hours in order to liberate any pyrimidine bases present. After removal of sulfates and phosphates and decolorization of the solution with charcoal, the neutral solu-

tion was concentrated to a small volume, which deposited crystals on long standing. The Wheeler and Johnson test for cytosine and uracil (9) was consistently negative when applied to these solutions or crystals. These results indicate that nucleic-acid derivatives are absent from the "alcohol product" or present only in very small quantities.

Isolation of an ash-free product from the "alcohol product"

Inasmuch as organic-phosphorus products obtained from soils have all contained inorganic matter, the possibility that the so-called "organic phosphorus" may consist of adsorbed inorganic phosphates is suggested. Subsequent experiments showed, however, that the mineral matter could easily be removed from the "alcohol product" by differential adsorption with charcoal, demonstrating that the phosphorus exists in true organic combination.

The "alcohol product" was dissolved in 5 per cent hydrochloric acid and vigorously shaken with a large quantity of phosphorus-free activated charcoal. The suspension was filtered, and the precipitate was washed with dilute hydrochloric acid and finally with distilled water until chloride- and phosphate-free. The filtrate and wash waters contained the inorganic materials, while most of the organic phosphorus remained adsorbed by the charcoal. The organic phosphorus was liberated from the charcoal by repeated extraction with dilute ammonium hydroxide. The extract was concentrated *in vacuo* to a small volume and allowed to stand in a vacuum desiccator over sulfuric acid until a thick sirup was formed. To separate solid material, the sirup was rubbed with absolute methanol, and the precipitate was centrifuged off, washed with small portions of absolute methanol, and dried to a tan-colored product. A product isolated in this manner from Honokaa soil contained 7.4 per cent phosphorus, all in organic combination, and 6.9 per cent nitrogen. Further purification was effected by successively preparing the barium and lead salts, and the final solution resulting from decomposing the lead salt with hydrogen sulfide was concentrated to a small volume under diminished pressure. Absolute ethanol was then added, and the amorphous precipitate formed was filtered off. The alcoholic filtrate was made alkaline with dilute ammonium hydroxide, thereby forming a second precipitate which was dried at 60°C. for several days. This product was soluble in water, very hygroscopic and ash-free, and contained 8.76 per cent phosphorus and 8.40 per cent ammonia nitrogen. Inasmuch as the yield of this material was very small, no attempt was made to purify it further; instead, it was reserved for hydrolysis.

Isolation of inositol

In previous attempts to isolate pyrimidine bases from the "alcohol product," it was observed that a small quantity of crystalline material could be obtained from the hydrolyzate. These crystals were nitrogen-free and later were identified as inositol.

Inositol from the "alcohol product." A 25-gm. portion of the "alcohol product" obtained from Honokaa soil was hydrolyzed with 100 cc. of 10 per

cent sulfuric acid (by weight) at 160°C. for 5 hours. The hydrolyzate was diluted with 300 cc. of hot water, filtered, and washed with hot water until the filtrate was colorless. Sulfuric and phosphoric acids were removed from the combined filtrate with barium hydroxide, and the excess barium was removed quantitatively with dilute sulfuric acid (1:20). The pale yellow solution was concentrated to a small volume, from which salts were precipitated by the addition of sufficient 95 per cent ethanol to make an 80 per cent ethanol solution. After salts were removed and the filtrate was evaporated to a volume of about 5 cc., 75 cc. of absolute ethanol was added and the solution was allowed to stand in the refrigerator for 24 hours. The crystals formed were separated on a sintered glass filter, washed with portions of absolute ethanol, and dissolved in 10 cc. of hot water. The aqueous solution was decolorized with a small quantity of charcoal (norite) and concentrated to about 5 cc. on the water bath. The addition of 50 cc. of 95 per cent ethanol immediately precipitated more inorganic salts, which were filtered off at once. After the filtrate stood at room temperature for 24 hours, impure crystals of inositol separated out. After four recrystallizations from absolute ethanol, about 100 mgm. of pure inositol was obtained with a melting point of 222° to 223°C. A mixed melting-point determination with pure inositol (Eastman) showed no depression. The crystals were soluble in water and insoluble in alcohol and ether. The Scherr-Salkowski color reaction (7) for inositol was positive. Elementary analysis showed the following composition: Found: C 39.52, H 7.14; calculated: C 39.98, H 6.67. Plate 1 shows a photomicrograph of inositol crystals obtained from Honokaa soil. Inositol was also isolated in pure crystalline form from the "alcohol products" of the Kona and University Farm soils.

Inositol from the ash-free product. Two-tenths gram of the ash-free product purified through the barium and lead salts and containing 8.76 per cent phosphorus was hydrolyzed with 2 cc. of 10 per cent sulfuric acid in a sealed tube at 160°C. for 5 hours. Crystals of inositol were obtained in sufficient quantity and purity for a melting-point determination of 221°C. The Scherr-Salkowski reaction was also positive.

Quantitative estimation of inositol from soil

In order to determine the extent to which inositol phosphates are present in soil, a quantitative method of estimating inositol from soil was developed by modifying slightly the methods of Young (12, 13). Briefly, the procedure consisted of the following operations: hydrolysis of the "alcohol product," isolation of crude inositol, and estimation of inositol by Young's modification of the Fleury-Marque iodomercurate method (13). The total amount of organic phosphorus hydrolyzed was calculated as the difference between inorganic phosphorus before and after hydrolysis. As a check on the accuracy of the method, a known quantity of inositol was hydrolyzed with the "alcohol product," and the amount of inositol recovered was determined. The results of the inositol determinations and their relation to organic phosphorus are

shown in table 2. The recovery of 87.7 per cent inositol from soil products agrees with recoveries from animal tissues reported by Young (12).

It may be seen from table 2 that the inositol-phosphorus ratios obtained from the three soils are nearly the same, approximately 0.3. A comparison of this figure with ratios calculated for phytin and for inositol monophosphate shows that a large part of the organic phosphorus in the "alcohol product" is not associated with inositol.

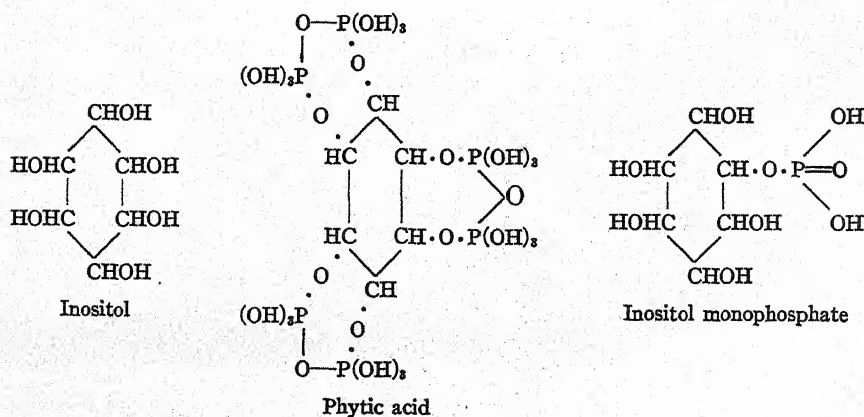
TABLE 2
Inositol and inorganic phosphorus produced by hydrolysis of the "alcohol products"

SOIL	INOSITOL*	PHOSPHORUS	INOSITOL/P RATIO FOUND	INOSITOL/P RATIO IN PHYTIN	INOSITOL/P RATIO IN INOSITOL MONO- PHOSPHATE
	mgm.	mgm.			
Honokaa.....	24.6	94.6	0.260	0.968	5.81
Kona.....	10.1	32.2	0.313		
University Farm.....	98.2	322.8	0.304		

* Maximum inositol values based on 87.7 per cent recovery.

DISCUSSION

The study of soil organic phosphorus in cultivated Hawaiian soils has brought out several outstanding points. The isolation of inositol establishes the presence of inositol phosphates in these soils and raises the question whether inositol is present as phytin. In considering the inositol-phosphate compounds of soil, a distinction should be drawn between phytin and other phosphate derivatives of inositol. Phytin is the calcium-magnesium salt of inositol hexaphosphoric acid (phytic acid), which apparently is easily susceptible to biological decomposition. On the other hand, Anderson (1) isolated crystalline inositol monophosphate from an organic-phosphorus fraction of wheat bran which had resisted breakdown into inorganic phosphates even after 2 years. The following formulas have been ascribed to inositol, phytic acid, and inositol monophosphate:



Anderson (2) also demonstrated that when phytase is not destroyed, all the phytin in wheat bran is completely changed within a short time, partly to intermediate derivatives such as inositol triphosphate and inositol monophosphate and partly to inositol and phosphoric acid. It is reasonable to believe that such changes may also take place when plant materials decompose in soil. Auten (3) conducted incubation experiments with phytin preparations and found that 67 per cent of the organic phosphorus was hydrolyzed to inorganic phosphorus at the end of 3 months. It was likewise probable that the organic phosphorus remaining after incubation was no longer due to phytin but to intermediate products derived from phytin. Dyer et al. (5) have suggested that phytin is present in soil as ferric phytate, which is not readily decomposed to inorganic phosphates. Conrad (4) has shown, however, that when phytin was added to a phosphorus-deficient soil, milo plants were benefited, indicating that the phosphorus of phytin becomes at least partly available. When plant materials decompose in soil, it is probable that phytin is converted to intermediate inositol phosphates before sufficient iron is available to precipitate ferric phytate.

The nature of the ash-free organic-phosphorus product isolated also suggests that inositol phosphates other than phytin exist in the soil. The properties of the ash-free product may be described as follows: resistance to decomposition; liberation of inositol and phosphoric acid on hydrolysis; and presence of 8.76 per cent phosphorus and 8.40 per cent ammonia nitrogen as compared with 10.54 per cent phosphorus and 9.52 per cent ammonia nitrogen as calculated for the ammonium salt of inositol monophosphate.

It has been shown from quantitative estimations of inositol that inositol-phosphate compounds compose only a part of the total organic phosphorus in soil. The approximate amounts of inositol-phosphate compounds in the "alcohol products" may be estimated by comparing the inositol-phosphorus ratios obtained with ratios calculated for phytin and for inositol monophosphate. The inositol-phosphorus ratios of the "alcohol product," of phytin, and of inositol monophosphate are roughly 0.3, 1, and 6, respectively. If it is assumed that all the inositol is derived from phytin, approximately 30 per cent of the organic phosphorus is accounted for. Only 5 per cent of the organic phosphorus in the "alcohol product" would be accounted for on the basis of inositol monophosphate. It must be remembered, however, that the "alcohol products" obtained from soil usually contain only about 50 per cent of the total organic phosphorus. In any case, since inositol phosphates do not contribute a large share to soil organic phosphorus, other compounds must be investigated in order to explain the apparent abundance of this bound phosphorus.

SUMMARY

In this study of the organic phosphorus in cultivated Hawaiian soils, an attempt was made to identify the organic-phosphorus compounds of soil. The results of the investigation may be summarized as follows:

Crystalline inositol was isolated for the first time from organic-phosphorus-bearing soil materials, thus establishing the presence of inositol phosphates in these soils.

No evidence of nucleic-acid compounds could be found in any of the soils investigated.

An ash-free organic-phosphorus product isolated from soil was resistant to decomposition and gave inositol and phosphoric acid on hydrolysis, and the phosphorus and nitrogen contents suggest the ammonium salt of inositol monophosphate.

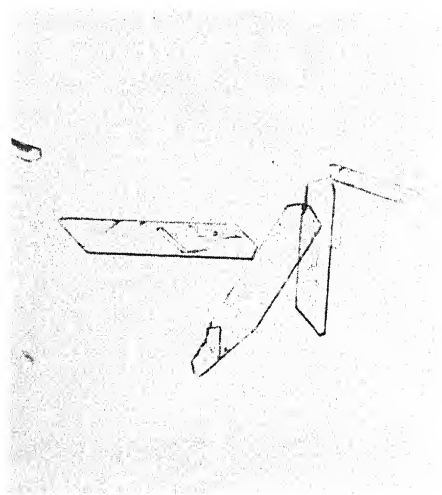
Quantitative estimations of inositol liberated by hydrolysis showed that inositol-phosphate compounds account for only a part of the total soil organic phosphorus.

REFERENCES

- (1) ANDERSON, R. T. 1914 Inosite monophosphate, a new organic phosphoric acid occurring in wheat bran. *Jour. Biol. Chem.* 18: 441-446.
- (2) ANDERSON, R. T. 1915 The hydrolysis of phytin by the enzyme phytase contained in wheat bran. *Jour. Biol. Chem.* 20: 475-482.
- (3) AUTEN, J. T. 1923 Organic phosphorus of soils. *Soil Sci.* 16: 281-294.
- (4) CONRAD, J. P. 1939 Retention of phosphorus compounds by soils. *Jour. Agr. Res.* 59: 507-518.
- (5) DYER, W. J., WRENSHALL, C. L., AND SMITH, G. R. 1940 The isolation of phytin from soil. *Science* 91: 319-320.
- (6) LEVENE, P. A., AND BASS, L. W. 1931 Nucleic acids. Amer. Chem. Soc. Monog.
- (7) SALKOWSKI, E. 1910 Kleinere Mitteilungen. *Ztschr. Physiol. Chem.* 86: 478.
- (7a) SCHREINER, O. 1923 Organic phosphorus in soils. *Jour. Amer. Soc. Agron.* 15: 117-124.
- (8) SHOREY, E. C. 1913 Some organic soil constituents. U. S. Dept. Agr. Bur. Soils Bul. 88.
- (9) WHEELER, H. L., AND JOHNSON, T. B. 1907 On a color test for uracil and cytosin. *Jour. Biol. Chem.* 3: 183-189.
- (10) WRENSHALL, C. L., DYER, W. J., AND SMITH, G. R. 1940 Recent studies on the nature of soil organic phosphorus. *Sci. Agr.* 20: 266-271.
- (11) WRENSHALL, C. L., AND MCKIBBIN, R. R. 1937 The nature of the organic phosphorus in soils. *Canad. Jour. Res. (B)* 15: 475-479.
- (12) YOUNG, L. 1934 The determination of inositol in animal tissues. *Biochem. Jour.* 28: 1428-1434.
- (13) YOUNG, L. 1934 The volumetric determination of inositol. *Biochem. Jour.* 28: 1435-1443.

PLATE 1

PHOTOMICROGRAPH OF INOSITOL ISOLATED FROM AN ORGANIC-PHOSPHORUS PRODUCT OF SOIL FROM HONOKAA, HAWAII



RELATION OF pH TO PHOSPHATE SOLUBILITY IN COLORADO SOILS

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The total phosphate supply in most Colorado soils is adequate to take care of crop needs for many years, but the predominant part of the phosphorus compounds is so slightly soluble or so firmly sorbed that the plants are often unable to get enough to supply their growth requirements. In order to maintain these soils in a high state of fertility, farming practices must be adjusted so that the soil phosphates can be more readily released to the plants, or the addition of phosphate fertilizer will be necessary. An intelligent control of the situation will not be possible until we are able to determine with reasonable accuracy whether or not the soil is deficient in readily available phosphorus and what effect the various farming practices have on phosphate availability. The problem is complicated by the fact that soils vary widely in composition from one locality to another, and these variations may greatly affect phosphate availability.

The soils in Colorado are generally well supplied with calcium carbonate and, therefore, tend to be alkaline in reaction. This condition leads to the assumption that most of the phosphorus not in organic combination is present in combination with calcium, only minor amounts being combined with iron or aluminum or sorbed by the colloidal silicates. The fact that calcium phosphates are readily soluble in acid but only slightly soluble in basic solutions suggests that the availability of these phosphates to plants in alkaline soils depends at least partly upon the acid supplied by either the plant roots or other biological processes.

In the absence of a thorough knowledge of the processes by which phosphate ions are transferred from the soil minerals to the plants, it may be assumed that either or both of the following means of entrance may be involved: intake from the soil solution, or ionic exchange between the colloidal fraction of the soil and the plant roots. If the solubility of the phosphate is very low within the possible pH range of the soil solution, a low availability can be expected unless there is at the same time an available supply of exchangeable phosphate ions attached to the colloid sufficient to meet the plant needs. Burd and Murphy (3) and Murphy (6) have shown that some soil colloids have the capacity to sorb a large quantity of phosphate which is not readily available to plants until a high percentage of the saturation capacity is satisfied. They show that in "strongly saturated" colloids some of the phosphate is readily

available. Whether it is made available through a process of ionic exchange between the plant roots and the soil colloid or is first taken into the soil solution is a question.

The most important factor which seems to determine whether Colorado soils can supply crops adequately with phosphorus is the solubility of the phosphates over the range of pH values of the soils in the vicinity of the plant roots. A second factor is the possibility of replacement of sorbed phosphorus from the colloidal complex. The most direct and most logical attack of the problem, therefore, seems to be the determination of, first, the solubility as a function of the pH; second, the pH range of the soil under field conditions; and, third, the sorbed phosphorus and the sorption capacity. With this information it should be possible to predict the need for phosphate fertilization and to determine the effect of farming practices on phosphate availability.

Accordingly, a study of these factors, together with a study of the probable pH feeding range of the plants and the total phosphorus content of the soils, is in progress. This paper is a report of results to date.

EXPERIMENTAL MATERIALS AND METHODS

Soils

Twelve soils representative of the plains, foothills, and mountain areas were selected for this report. A brief description of them is given in table 1.

Solubility and buffer curves

Soil samples of 4 gm. each were treated in large test tubes with 40 cc. of sulfuric acid or potassium carbonate of various concentrations to produce the desired pH range. These reagents were chosen because the most important soluble bases in alkaline soils occur as salts of carbonic acid (CO_3^{--} and HCO_3^-) and because the soluble salts in Colorado soils are predominantly sulfates. The suspensions were shaken for an hour and allowed to settle overnight. The supernatant liquid was analyzed colorimetrically for phosphate without treatment for the decomposition of organic matter except that colored solutions were treated with bromine. The pH of the solutions was determined with a glass electrode.

pH at the moisture equivalent

Samples of soil at the moisture equivalent were placed in moist chambers filled with CO_2 and allowed to stand for about 72 hours. A similar set of samples was placed in chambers with air instead of CO_2 . At the end of this time, the pH of the moist soil was determined by inserting a glass electrode into the soil.

Sorbed phosphorus and sorption capacity

The method of Burd and Murphy (3) was used to estimate sorbed phosphorus and sorption capacity, except that 2 per cent KCl solution instead of water

was used to remove the excess phosphate and the soil solution ratio was 1:10 instead of 1:5. Determinations were also made with a 1 per cent K_2CO_3 solution instead of 0.1 *N* NaOH.

Total phosphorus

Total phosphorus was determined by the magnesium nitrate method of the Association of Official Agricultural Chemists (1).

TABLE 1

Colorado soils studied

SOIL NUM- BER	DEPTH	P ₂ O ₅	CaCO ₃ *	SOIL TYPE	AVAILABLE P	REMARKS
	<i>inches</i>	<i>per cent</i>	<i>per cent</i>			
1	0-6	.21	4.8	Fort Collins loam	High	Very fertile
2	0-6	.19	6.9	Fort Collins loam	Moderately de- ficient	Very fertile if phos- phated
3	0-6	.26	9.2	Unclassified loam	High	Chlorotic peach or- chard
4	12-24	.23	26.6	Unclassified clay	Not known	Subsoil of No. 3
5	0-6	.28	9.7	Billings clay	Not known	High alkali, not productive
6	0-6	.18	4.5	Weld fine sandy loam	Very deficient	"Black heart" sugar beet field
7	0-6	.14	4.5	Larimer fine sandy loam	Very deficient	"Black heart" sugar beet field
8	0-6	.16	5.8	Weld fine sandy loam	Very deficient	"Black heart" sugar beet field
9	0-6	.19	2.8	Cass fine sandy loam	Moderately de- ficient	Responds to phos- phate with beets
10	0-6	.12	0.0	Neville fine sandy loam	Well supplied	Does not respond to phosphate with cherries
11	0-6	.14	0.0	Unclassified sandy loam	Very deficient	Will not produce crop without phosphate
12	0-6	.19	0.0	Unclassified muck	Not known	A mountain bog soil

* Total carbonate calculated as calcium carbonate.

Correlation between phosphate extracted by acids and bases

Phosphate extractions by the buffered acetate method of Dahlberg and Brown (4), by the buffered sulfuric acid method of Truog (8), by 0.06 *N* H_2CO_3 solutions, by water saturated with CO_2 at 600 mm. pressure, by the potassium carbonate method of Hockensmith, et al. (5), and extractions from sugar beet petioles were checked for degree of agreement by calculating their correlation coefficients.

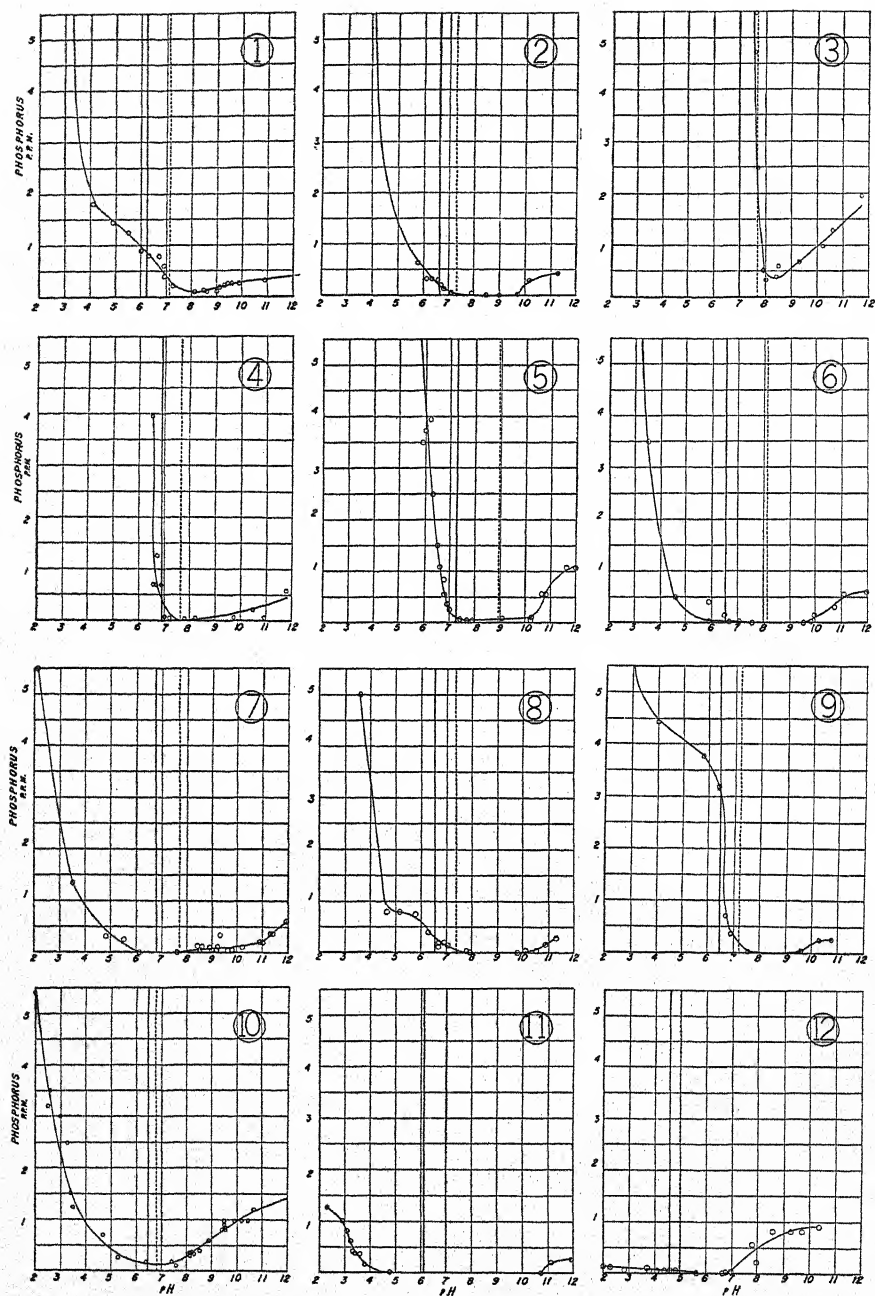


FIG. 1. EFFECT OF pH ON THE SOLUBLE PHOSPHORUS IN 12 COLORADO SOILS, EXPRESSED AS PARTS PER MILLION PHOSPHORUS IN SOLUTION

The heavy vertical lines show the pH of the soils at the moisture equivalent. The solid line at the left is the pH with the soil under 600 mm. CO₂ pressure, and the dotted line to the right, under air.

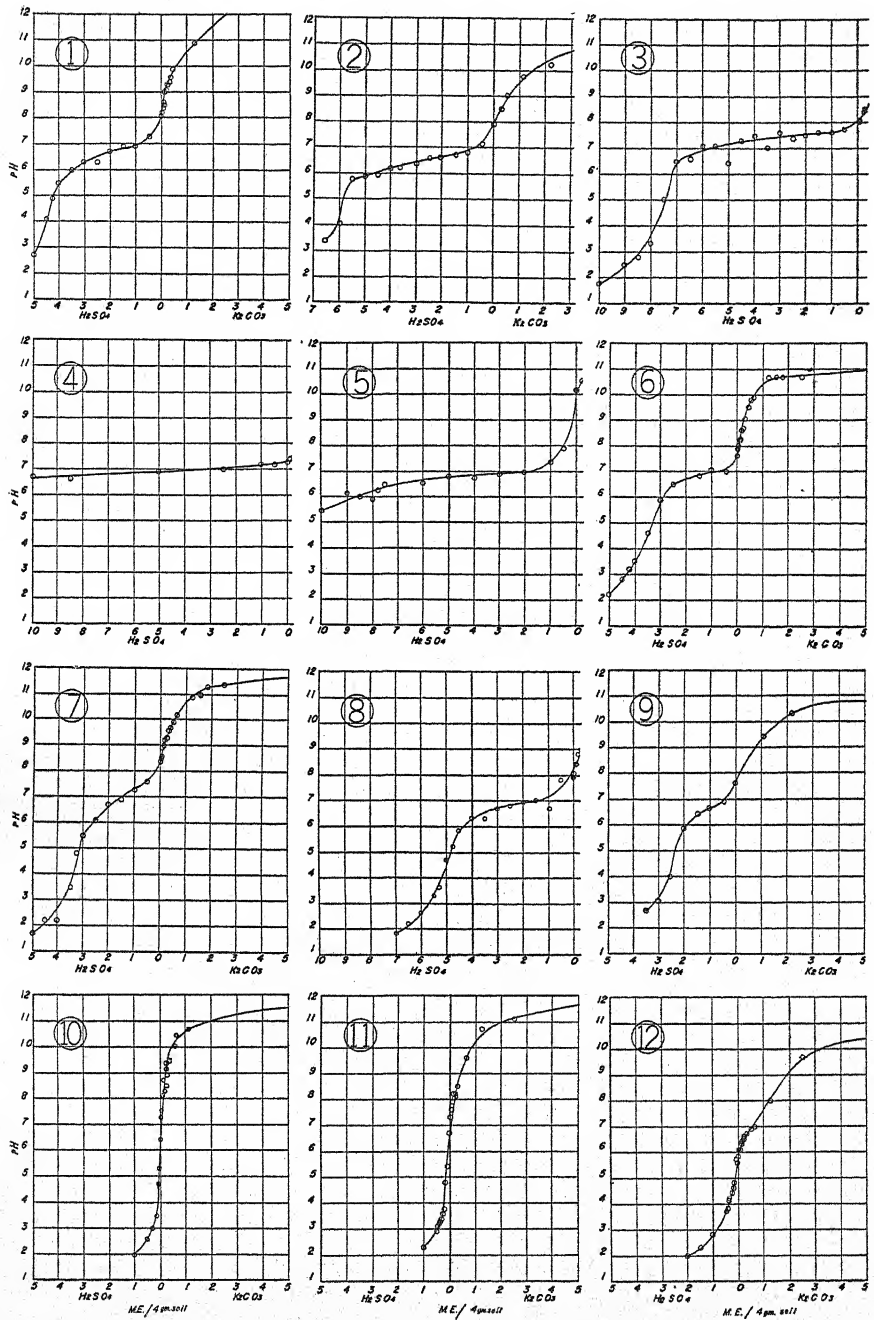


FIG. 2. EFFECT OF ACID OR BASE IN MILLIGRAM EQUIVALENTS PER 4 GM. OF SOIL ON pH OF THE 12 SOILS IN FIGURE 1

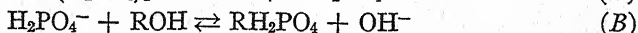
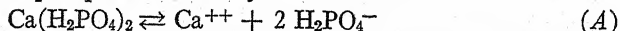
EXPERIMENTAL RESULTS

The phosphate solubility is shown graphically in figure 1, and corresponding buffer curves are shown in figure 2. Table 2 shows the amount of phosphorus in NaOH and K_2CO_3 extracts before and after saturation of the soils with phosphate. Table 3 gives the correlation coefficients between different extraction methods.

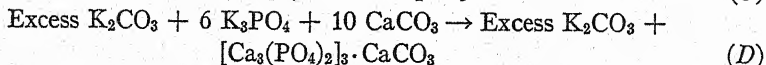
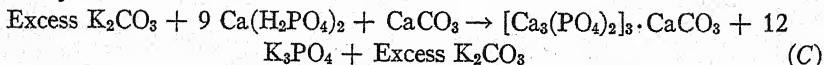
DISCUSSION

The phosphorus solubility curves in figure 1 differ widely but are all of the same general type with a minimum near the neutral point. The fact that there is a rise on the alkaline side indicates that other compounds than tribasic calcium phosphate are present. According to the work of Teakle (7), Buehrer (2), and others, the solubility of calcium-saturated phosphates should decrease with increasing pH on the alkaline side, and the work of Buehrer (2) shows that at equilibrium in the presence of excess calcium carbonate and sodium or potassium carbonate, a very slightly soluble carbonato-apatite should be formed. It is not likely, however, that the curves in figure 1 represent equilibrium conditions, and, therefore, at least part of the phosphate in solution might be from monobasic or dibasic calcium phosphates present before treatment. The strongly alkaline solutions would also dissolve iron or aluminum phosphates and displace phosphate ions from the colloidal clay. (The possibility that the rise on the alkaline side was due to error from the solution of silica in the alkaline extracts was investigated, but the results were not significantly changed after the extracts had been dehydrated four times with hydrochloric acid.)

Merely to illustrate in a simple form the type of reactions which could be expected, the following suggestive equations are given. Equations (A) and (B) indicate the equilibrium between monobasic calcium phosphate, the phosphate ions, and the phosphate sorbed by the colloid.

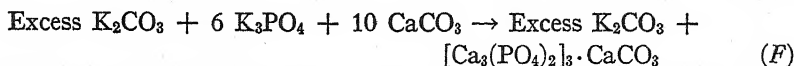
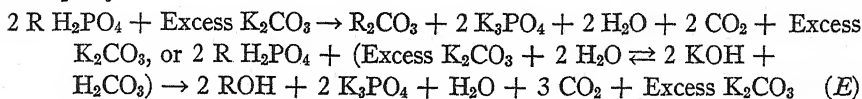


where R represents the colloidal particles. Equations (C) and (D) illustrate the effect on the unsaturated calcium phosphates of adding an excess of K_2CO_3 to the system.



Assuming that equation (D) represents the final reaction, the low concentration of calcium in solution due to the excess K_2CO_3 would greatly slow up the reaction beyond the steps represented in (C), resulting in an apparent equilibrium before actual equilibrium had been reached. It is also probable that at the pH of the suspensions the soluble phosphates would be partly dibasic rather than completely tribasic as the equations read, which would further retard the formation of carbonato-apatite.

The situation with respect to the phosphate sorbed by the colloid is similar to that of the phosphate in the unsaturated calcium phosphates in the presence of K_2CO_3 .



In this case, equation (E) would represent an apparent equilibrium, even though the reaction proceeded slowly, according to equation (F).

It is not intended to infer that the foregoing equations show the exact steps in the formation of carbonato-apatite, but, on the assumption that equilibrium tends toward the formation of some such very slightly soluble compound, they are offered as a possible explanation of the fact that the addition of K_2CO_3 increases the phosphate in solution.

In general, the rapid rise of the curves on the slightly acid side shows the presence of comparatively large quantities of calcium phosphate. The very low values in the slightly alkaline range where iron and aluminum phosphates are appreciably soluble indicate the nearly complete absence of these compounds. The rise in the more alkaline range shows that some phosphate either adsorbed to the colloid or as monobasic or dibasic calcium phosphate is likely present.

The heavy vertical lines in figure 1 show the pH of each soil at the moisture equivalent under CO_2 and under air. The CO_2 was held at 600 mm. pressure ± 30 mm. for about 72 hours. This is about the maximum pressure which could be obtained as the result of CO_2 given off from the plants, even in the immediate vicinity of the roots and, therefore, the pH should be the minimum average which could result from CO_2 , though lower values could conceivably result in local areas if these were free from $CaCO_3$. The normal pH of the soil should fluctuate somewhere between the two lines, depending on the CO_2 pressure. The solubility of phosphorus between these two lines, though not the true solubility under field moisture conditions, may indicate the relative ability of the soils to supply phosphorus to plants through the soil solution. The phosphate content on the extreme alkaline side serves as an estimate of the maximum amount available for feeding by ionic exchange between the plant and the soil colloid.

If the phosphorus on the extreme alkaline side of the curves in figure 1 is assumed to be the total phosphorus sorbed by the soil colloids in an exchangeable condition, it is evident that in the calcareous soils this supply would constitute only a very small reserve of available phosphorus, even if it were all readily available to the plants. To be an important factor in plant feeding, it would need to be replenished continuously from the ions in solution. Thus, the colloid would be either a source of supply to the plants or a competitor of the plants, depending upon the concentration in solution and the degree

of colloid saturation. The practical significance of this fact is that where a high saturation capacity is indicated in the presence of a low concentration in solution, it is likely that a large application of fertilizer would be required to bring the available supply up to the plant needs because of the competition of the colloid for the ions in solution. In a soil with a low saturation capacity more of the added phosphate would be available for the plants. This fact is verified by the work of Burd and Murphy (3) and Murphy (6).

Table 2 shows an estimate of the sorbed phosphorus and the sorption capacity plus any phosphorus from calcium or other phosphates as determined by treating the soil with NaOH and K_2CO_3 solutions before and after saturation

TABLE 2

Alkali-soluble phosphorus and alkali-soluble capacity of the soils as determined by treating with K_2CO_3 and NaOH before and after saturating with phosphate

SOIL NUMBER	BY NaOH			BY K_2CO_3		
	Alkali soluble	Alkali- soluble capacity	Degree of saturation	Alkali soluble	Alkali- soluble capacity	Degree of saturation
	<i>p.p.m. in solution</i>	<i>p.p.m. in solution</i>	<i>per cent</i>	<i>p.p.m. in solution</i>	<i>p.p.m. in solution</i>	<i>per cent</i>
1+	0.30	11.9	2.5	1.12	15.0	7.5
2-	?	20.7	Very small	0.53	20.2	2.6
3+	0.70	8.6	8.1	1.79	8.8	20.3
4*	0.30	1.5	20.0	0.45	50.5	0.9
5*	0.70	6.8	10.3	1.12	11.5	9.7
6-	0.60	14.4	4.2	0.56	14.4	3.9
7-	0.50	11.6	4.3	0.45	16.4	2.7
8-	?	14.4	Very small	0.42	14.4	2.9
9-	?	10.5	Very small	0.62	15.0	4.1
10+	1.70	15.0	11.3	1.72	14.0	12.3
11-	0.30	50.5	0.6	0.45	48.6	0.9
12*	95.9		1.05	70.7	1.5

+ = Well supplied with available P.

- = Deficient.

* = Unknown.

? = Concentration too low to be read.

with phosphate. It will be noted that the percentage saturation as estimated from the K_2CO_3 extracts agrees very closely with the available phosphorus: the soils known to be well supplied with phosphorus have the highest percentage saturation, and the ones known to be deficient have a low percentage saturation. A high correlation is also evident between the percentage saturation and the phosphorus in solution at the normal pH, shown in figure 1. The agreement was not so good where NaOH was used. This probably was because a large amount of soluble calcium was present in some soils, as in soil 4, for example, which contained calcium sulfate. The phosphate displaced apparently was precipitated as the calcium salt. Since the carbonate reduces

the calcium ion to a low point, it seems preferable to NaOH as a replacing agent for estimating adsorbed phosphorus in high-calcium soils.

Soils 1 to 9 are calcareous. The amount of lime is shown in table 1 and by the buffer curves in figure 2. Crops grown on soils 1 and 3 are well supplied with phosphorus; 2 and 9 are moderately deficient; and 6, 7, and 8 are very deficient. If the pH range between the two heavy vertical lines in figure 1 is considered as the approximate range at optimum field moisture, the soluble phosphorus in this range indicates the concentration available to plants in solution. Number 1 shows a moderate concentration, and No. 3 a high concentration, between the two lines, indicating that an appreciable supply would be available from solution. On the contrary, Nos. 6, 7, and 8 show a very low concentration in the normal pH range and also show that if the plants were able to reduce the pH below this range in local areas, these areas would need to be very acid to liberate an adequate supply. The low percentage saturation shown in table 2 also indicates a low available supply.

Soil 9 is moderately deficient for sugar beets but shows a very high phosphate content under 600 mm. pressure CO_2 . At the pH in air, with no addition of CO_2 , the phosphate content is low. This fact indicates that sugar beets planted on this soil do not change the pH so much as does an atmosphere of CO_2 and seems to discredit the idea that feeding by reducing the pH to a very low point in local areas is important. An atmosphere of CO_2 should be able to provide an abundance of phosphorus. Any extractant which gave a pH below 6.5 would indicate plenty of phosphorus, yet crops on this soil respond to phosphate fertilization. Soil 2 is not so striking in this respect as is No. 9, but the graph indicates that there should be a liberal supply slightly below the pH resulting from one atmosphere of CO_2 .

Soil 4 is the subsoil of 3 and gives a very similar curve except that the solubility of the former is very low over the pH range between the vertical lines. No data are available to indicate whether or not this soil will respond to phosphates. The curve is presented to show the importance of selecting the right extractant in estimating available phosphorus on soils of this type. Because of the steep slope, a slight shift one way or the other would determine whether a very little or a very large amount was brought into solution. If it is assumed that the phosphate under the heading "alkali-soluble capacity" in the K_2CO_3 -treated soil in table 2 is the phosphate sorbed by the colloid, this soil appears to have a high sorption capacity. It appears more likely, however, that much of this phosphate is from monobasic or dibasic calcium phosphate precipitated because of the calcium sulfate present in this soil during its treatment with phosphate solution.

Soils 10, 11, and 12 are all noncalcareous. Soil 10 is moderately productive. Cherries do not respond to phosphate on this soil. The curve is presented to illustrate the tendency to a higher value on the alkaline side and the shift of the minimum toward the acid side in noncalcareous soils. Soil 11 is so low in available phosphorus that it requires fertilization to produce any crop. Vir-

tually no phosphorus is in solution in any part of the possible feeding range of crops. It has a high saturation capacity and a low percentage saturation. Large applications of phosphate would likely be required to make this soil productive. Soil 12 is an acid muck from a mountain bog of unknown response to phosphate fertilization. The graph indicates that it is undoubtedly deficient, since there is virtually no phosphate in the acid range. It has a high saturation capacity and a low percentage saturation.

The data in table 3 show how closely results of phosphate tests of Colorado soils made during the last few years by several methods are correlated with

TABLE 3
Correlation of phosphate dissolved by various extracting solutions

SOIL GROUP NUMBER	LOCATION	NUMBER OF FIELDS	NUMBER OF PLOTS	NUMBER OF PAIRS OF SAMPLES	DEPTH SAMPLED	METHODS CORRELATED	CORRELATION COEFFICIENT	
							Separate samples	By depths*
					<i>inches</i>			
1	Distributed over Colo.	89		89	0-6	K ₂ CO ₃ and acetate	.64	
2		89		89	0-6	K ₂ CO ₃ and Truog	.47	
3		89		89	0-6	Acetate and Truog	.59	
4		35		89	0-6	K ₂ CO ₃ and 0.06 N CO ₂	.88	
5	Agronomy Farm	1	24	96	0-36	K ₂ CO ₃ and acetate	.79	.97
6		1	84	84	0-6		.72	
7	Loveland	1	18	72	0-36		.79	.96
8	Fort Collins	1	32	128	0-36		.77	.98
9	Masonville	1	24	96	0-36		.67	.94
10	Loveland	1	18	72	0-36		.54	.63
11	Palisades	9	18	126	0-72		.78	.99
12	Agronomy Farm	1	84	84	0-6	K ₂ CO ₃ and atmosphere CO ₂	.73	
13		1	84	84	0-6	K ₂ CO ₃ and P in beet petioles	.867	
14		1	84	84	0-6	Acetate and P in beet petioles	.871	

* Correlation coefficients of the mean value of each of the following depths: 0-6, 6-12, 12-24, and 24-36 inches.

each other. The data are reported because they give additional information on the behavior of the soil phosphates as affected by the pH of the extracting solution and also because they throw some light on the suitability of various extracting agents in soil testing. The pH of the Truog reagent was 3.0; that of the acetate reagent, 4.9; and that of the K₂CO₃ solution, above 12. The exact pH of the two CO₂ solutions used is not known.

The fact that there is a significant correlation between the results of any two of the methods indicates that the solubility curves tend to parallel each other through the pH range resulting from the various extractants. With

one exception, the lowest correlation coefficients were obtained when the results from the Truog test were one of the variables in the correlation. This fact could be due to a slighter tendency for the curves to run parallel in the more acid range or to a lower buffering capacity of the Truog extractant, or both. The close correlations between the phosphate in sugar beet petioles grown on the soil and the acetate and K_2CO_3 solution extracts of the soil indicate that extracts with these reagents of soils similar to those on the Agronomy Farm are closely correlated with plant available phosphorus. The correlation between the results of these two extractants is very close when there is not a great variation in soils, but the correlation decreases when a wide variety of soils is used, as is indicated in the results of the first group correlated. This would be expected if the solubility curves in figure 1 are studied. These curves show that in the noncalcareous soils the minimum tends to shift toward the acid side, and the alkaline side tends to be higher. For this reason, the K_2CO_3 method would tend to indicate plenty of available phosphorus in noncalcareous soils if standardized with calcareous soils, and the reverse would be true of the acetate method.

Insofar as the data presented will justify generalization, it appears that curves showing the effect of pH on solubility are capable of furnishing the basis of a very good prediction of the probable need for phosphate fertilization in alkaline soils. The preparation of such graphs is probably too laborious to meet the needs of routine soil testing where the so-called "quick tests" have come into wide use, but they offer a basis for the selection of appropriate extractants for use in the "quick tests" and show some of the limitations of these tests. No attempt will be made to draw accurate conclusions at this time, but attention is directed to the following indications:

The slopes of the curves on the acid side are so variable that any extractant which might reduce the pH very far below the normal field reaction would likely give erratic results poorly correlated with available phosphorus.

The buffer capacity of the soils is so variable that a fixed quantity of a strong acid sufficiently concentrated to reduce the pH to field conditions in some soils would reduce it too far in others. A strong acid, therefore, does not seem appropriate for Colorado soils unless the amount used is varied according to the buffer capacity of the soil.

An alkaline extractant such as the potassium carbonate solution of Hockensmith, et al. (5), seems to owe its value to the fact that with soils of similar composition there is a correlation between the alkaline- and acid-extractable phosphorus. The method, though it gives fair results in Colorado, is evidently not adaptable where the soils vary widely in composition and especially if noncalcareous soils are included with calcareous soils, unless the alkali-soluble phosphate is considered in relation to the sorption capacity.

A pure water extractant seems unsuitable as a means of determining available phosphorus, because the dilution with water tends to raise the pH so much in many suspensions that it is far above the range at normal moisture and because the solutions tend to be turbid. Comparison of the pH at the moisture equivalent on the phosphate curves with that on the buffer curves at the point where no acid or base was added will show the change in pH due to dilution. It will be noted that only soil 5 is alkaline in 600 mm. CO_2 at the moisture equivalent.

Since response to phosphate fertilization depends upon the capacity of the soil to sorb

phosphorus as well as upon the solubility of the soil phosphates, it is not likely that any acid extractant will give wholly accurate indications of plant response to phosphate fertilization unless something is known of the degree of saturation and saturation capacity of the sorption complex.

Alkali extractions before and after saturation of the soil with phosphate are capable of furnishing a basis for an estimate of the degree of saturation and the saturation capacity of the sorbing complex.

SUMMARY

Data have been presented on the effect of pH on phosphate solubility in some Colorado soils. These data show the following trends:

The solubility reaches a minimum near the neutral point.

The solubility in the pH range corresponding to the probable range at field moisture appears to be closely related to plant available phosphorus.

The solubility in the slightly acid range is closely correlated with solubility in the highly alkaline range for soils of similar composition.

The solubility in the probable range of pH at field moisture is correlated with the degree of colloid saturation as estimated by methods similar to that of Burd and Murphy (3).

Some practical applications of the data are as follows:

They can serve as an estimate of whether crops will respond to fertilizer on the soils studied.

They indicate the general type of extractants suitable for quick tests of phosphate availability.

They offer a possible means of estimating the amount of phosphate from fertilizer applications which will be adsorbed by the soil in a form not readily available to plants.

REFERENCES

- (1) Association of Official Agricultural Chemists 1930 Official and Tentative Methods of Analysis. Washington, D. C.
- (2) BUEHLER, T. F. 1932 The physico-chemical relationships of soil phosphates. Ariz. Agr. Exp. Sta. Tech. Bul. 42.
- (3) BURD, J. S., AND MURPHY, H. F. 1939 The use of chemical data in the prognosis of phosphate deficiency in soils. *Hilgardia* 12: 323-339.
- (4) DAHLBERG, H. W., and BROWN, R. J. 1932 A study of the Neubauer and Winogradsky (azotobacter) methods as compared with a chemical method for the determination of phosphorus deficiency in western soils. *Jour. Amer. Soc. Agron.* 24: 460-468.
- (5) HOCKENSMITH, R. D., GARDNER, R., AND GOODWIN, J. 1933 Comparison of methods for estimating available phosphorus in alkaline calcareous soils. Colo. Agr. Exp. Sta. Tech. Bul. 2.
- (6) MURPHY, H. F. 1939 The role of kaolinite in phosphate fixation. *Hilgardia* 12: 343-382.
- (7) TEAKLE, L. J. H. 1928 Phosphate in the soil solution as affected by reaction and cation concentrations. *Soil Sci.* 25: 143-162.
- (8) TRUOG, E. 1930 The determination of the readily available phosphorus of soils. *Jour. Amer. Soc. Agron.* 22: 874-882.

SOIL PROFILES IN RELATION TO THE RECESSION AND EXTINCTION OF MICHIGAN LAKES¹

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The number of lakes in Michigan is not definitely known but very probably exceeds 5000, if all kinds are included. The number of extinct lakes, or basins which formerly contained water, may be several times this figure, and the aggregate surface area of water in the basins of existing lakes is probably less than half that represented by old shore lines.

Recession of shore lines and extinction of lakes have been the rule in the history of the land surface. The probable causes are changes in climate; natural drainage by streams; and filling by aquatic vegetation, by accumulation of marl, or by sediment carried by inflowing streams or washed from the slopes of the enclosing basins. Also some recent change has been brought about by artificial drainage and clearing of land for agricultural use. The relative time at which changes may have taken place, the relative permanency of different lake types, and the present rate of recession and extinction, are of economic as well as of purely scientific interest.

A study of the soil profiles of lakes and of former lake beds in Michigan was made in the expectation that such a study might aid in the formulation of explanations and theories for lake extinction and other phenomena of change. Inferences from pedologic data may be of value because of the known relations of the soil profile to the depth of water table, to water conditions under which the soil has developed, to vegetation, to age of the land surface, and to the character of the geologic parent material.

A series of generalized soil profiles from water to a very dry condition is illustrated in figure 1. This constitutes a soil catena, represents the stages of soil development, and at least roughly comprises a series arranged according to age.

The water soils, or hydrosols, have not yet been satisfactorily classified. They are so diverse in details of chemical and physical character that a list of all possible kinds cannot be presented in any taxonomic scheme on the basis of present knowledge and observations. The subaqueous horizon of the soil profile, or bottom deposit, can be divided, however, into two general kinds of material: organic and inorganic.

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The organic bottom deposit is not uniform in chemical composition or even in color; it is also variable in texture and consistency from a fluid slime to a fairly compact mud, or it may be coarse, fibrous, and matted. After exposure, the degrees to which this material has been altered in structure, leached of its more soluble constituents, and humified may be used as a rough measure of time since recession or disappearance of the water. Given sufficient time, the lake bottom soil will completely lose its original character and be transformed into a mature mineral soil profile.

The inorganic type of bottom varies widely in chemical composition and ranges from a sticky colloid to coarse sand and gravel, or may consist largely of marl. Under water, the inorganic matter is usually covered by a thin film of slimy organic matter, with a variable amount of which it is mixed. Sands,

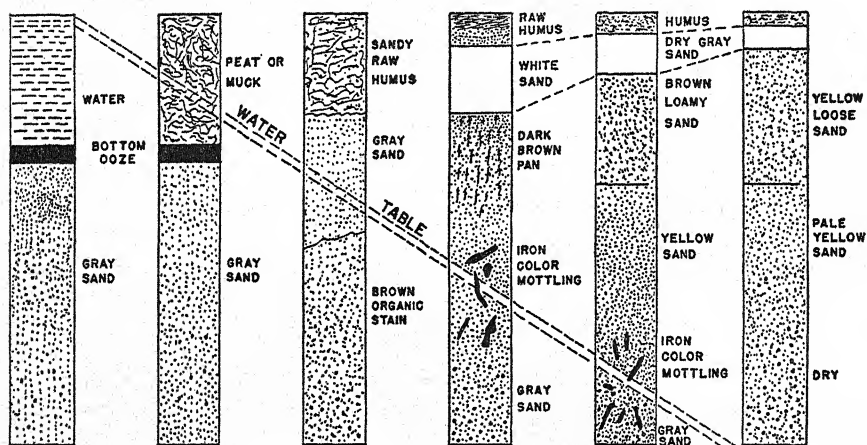


FIG. 1. SERIES OF SOIL PROFILES FROM WATER TO DRY SAND

The age of the soil and of the land surface which it represents increases from left to right

while still covered by shallow water, in places exhibit a weak soil profile apparent as a faint brown organic stain a few inches to about a foot from the surface, or as iron oxide coloring of some yellow shade. When the lake bottom becomes dry land, the sands are rapidly leached, lose the organic matter of lacustrine origin, and become gray or white at the surface; or eventually they may become covered with humus from dry land vegetation, develop a highly podzolized horizon, take on yellow ferric oxide coloring at depths, and assume a great number of variations in profile depending upon the depth to the water table, age, and other factors. The more argillaceous bottoms, after becoming dry land surfaces, rapidly develop a black humus covering, especially if they are calcareous; a gray, green, or bluish-gray glei horizon; and a yellow iron oxide staining, or mottling, at lower depths. Soluble constituents are removed much more slowly than in sands; and therefore removal of free calcium

carbonate, for example, to depths of 2 or 3 feet would require a relatively long time. The nearly pure marls change very slowly.

The changes that lakes have undergone since their birth are exceedingly diverse and complicated. Each lake is a separate entity, and it is scarcely conceivable that any two are exactly alike in origin and morphologic character,

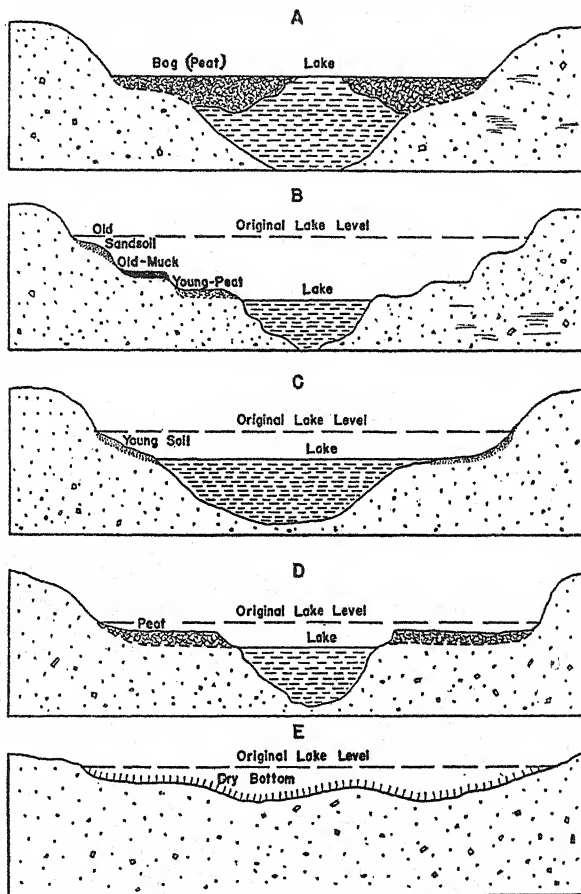


FIG. 2. TYPES OF RECESSION AND EXTINCTION OF LAKES

A, filling without drop in water level; B, slow recession by separate stages; C, rapid recession without filling by vegetation; D, recession after partial filling by vegetation; E, complete extinction of very shallow lake due to abrupt drop in water level

or that changes which have taken place in lakes and lake basins are precisely the same for any two. It may be possible, however, to group the changes into broad types, some of which are illustrated in figure 2. Some lakes have undergone a long slow recession by definite stages, as indicated by wide zones or several different kinds of soils in the basin as illustrated in figure 3. On the

other hand, some have undergone, and are still undergoing, a rapid continuous recession, as indicated by the uniformity of the soil profile in the former bed (pl. 1, fig. 1). By inference, the water in certain basins has disappeared suddenly and completely, since the soil is uniform or in the same stage of profile development throughout the basin. In many instances (pl. 2, fig. 1) disappearance of the water has been so recent that the soil is still essentially a

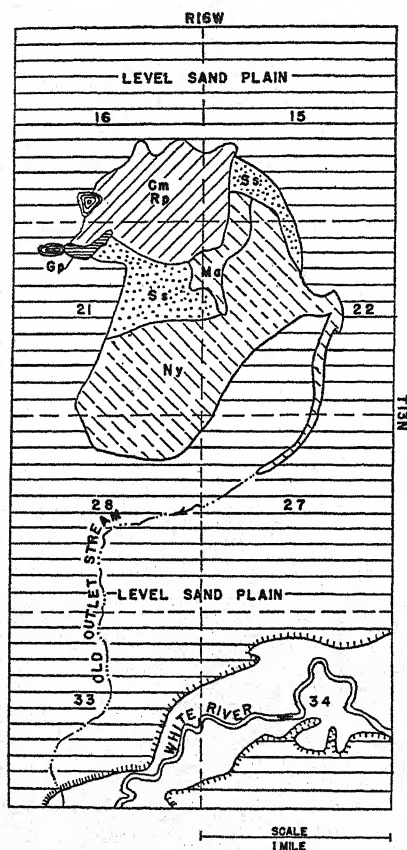


FIG. 3. SWAMPY BASIN ONCE THE SITE OF A LARGE LAKE

The successive stages of extinction and the ages of the soils from oldest to most recent are indicated by the symbols Ss, Ny, Ma, Cm, Rp, Gp (From Soil Survey of Oceana County, Michigan, U. S. Dept. Agr. Ser. 1933, No. 12, Wonser, C. H., Veatch, J. O., and Jones, L. R.)

hydrosol; and, on the other hand, in many, the extinction must have taken place during an early postglacial period, since the soil closely approaches the mature, or zonal, profiles of this region. The implication is that if the character of individual profiles in the series for all the possible catenas of a particular region is known, the relative time since lake recession or complete extinction took place may be inferred by comparison of the soil profile in the lake basin with standard profiles.

Basins which have a soil profile indicative of marsh or bog conditions but which are now so dry that the water table is 3 to more than 5 feet below the surface are common in parts of Mason, Newaygo, Oceana, and Muskegon Counties. In places, former shallow grass or marsh lakes changed to prairies characterized by a thick surface layer of dark colored soil underlain by sand and gravel, dry to a depth of several feet. Soils of this origin have been classified in Michigan as the Sparta series by the Division of Soil Survey of the U. S. Department of Agriculture.

Filling by aquatic, or hydrophytic, vegetation (pl. 1, fig. 2) is probably the most common single cause of lake extinction. Filling in all stages of completion may be observed at the present time, and destruction of lakes from this cause was probably quite as common in the past, since very old organic soils and forested swamps are found in basins which were evidently once occupied by bodies of water. In the soil mapping in the state a number of types of organic soils have been recognized which can be arranged in a series according to the degree of change in the parent organic material.

The stage of soil profile development, of course, cannot be correlated precisely with age, but in general there must be a measure of agreement. For example, given two areas both representing the former bottoms of separate lakes, or separate portions of the same lake, if the soil of one area is properly classified as Carlisle muck and the soil of the other as Greenwood peat, it may be safely assumed that the first named soil represents an older land surface.

On the basis of general field observations, coupled with a study and interpretation of the more detailed soil maps of the state, the conclusion has been reached that extinction of lakes is not a phenomenon of recent occurrence. Probably this has been the general trend since the final retreat of the ice sheet, although doubtless there were periods of fluctuation in water levels. As evidence that some lakes disappeared early in the postglacial period, the present soil profile in the basins closely resembles that of the adjacent upland.

The mortality of lakes apparently has been highest for the shallow pit, or saucer-basin, types. The deep lakes exhibit a higher survival and less recession than the shallow ones, although a few exceptions have been noted, as for example in deep lime sinks in Presque Isle and Alpena Counties. Likewise, basins filled to a depth of 60 to 70 feet by peat have been found.

In relation to physiographic divisions of glacial origin, the greatest number of waterless depressions appear on the outwash plains of sand and gravel, although on some of the rolling till clay plains the percentage of depressions may be quite as great. There appears to be no uniformity of survival or mortality, since living lakes, exhibiting little or no recession and in an apparently young state of development, may be closely associated with extinct ones, in the same topographic and geologic environment. An illustration of this is given in figure 4.

Again, no very apparent difference exists between the age of the land surface and the proportional number of extinct lakes and amount of filling. The southern part of the state presumably is older than the northern part, since

the ice retreated earlier there, but from inspection of soil maps throughout the state no very great differences are apparent. Also, filling and extinction may be observed on all lithologic types of glacial drift and seemingly bear little relation to the thickness of the drift.

The theory that lakes and swamps on an elevated and young glaciated surface such as Michigan are relatively ephemeral is generally accepted by geologists and physiographers. Pedologic evidence amplifies other kinds of evidence that there is a persistent trend toward extinction due to natural causes. There are a few instances, however, of a rise in water level at present, and marked fluctuations in the past may be inferred. A few instances of new

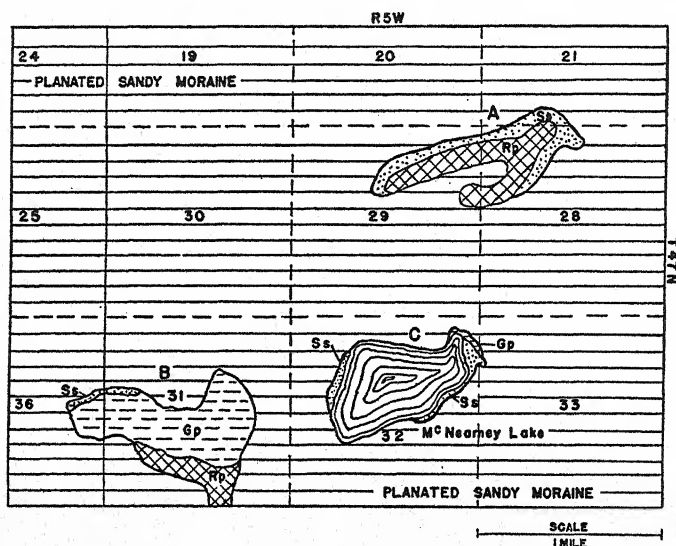


FIG. 4. DIFFERENT STAGES OF EXTINCTION IN THREE LAKES IN THE SAME GEOLOGIC ENVIRONMENT

A, first to become extinct; B, very recent extinction; C, a young lake, very little recession (From Soil Survey of Chippewa County, Michigan, U. S. Dept. Agr. Ser. 1927, No. 36, Veatch, J. O., et al.)

lakes have also been noted. For example, a small natural lake in the northern part of Marquette County is of such recent origin that some of the pine trees which formerly grew in the dry basin are still standing. If soil profiles in lake basins are scrutinized in detail, they will show traces of any changes in water levels which were of considerable duration. For example, from a study of the soil profile pictured in plate 2, figure 2, it may be inferred that the land was sufficiently inundated to enable a foot or so of finely fibrous Houghton peat to accumulate on an old soil profile of a series (Saugatuck) which is believed to have developed on sand parent material with a water table 2 to 4 feet from the surface.

PLATES

PLATE 1

FIG. 1. Recent rapid recession of a lake level. The stages are indicated by the numerals. The soil of the exposed lake bed is very young. Thomas Lake, Otsego County, Michigan

FIG. 2. Lake showing recent rapid filling by vegetation, followed by a drop in water level. Watkins Lake, Jackson County, Michigan



FIG. 1

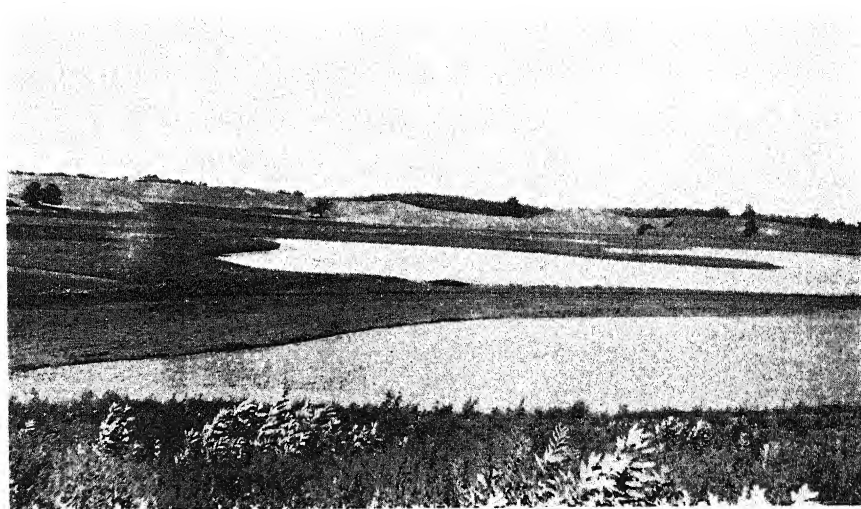


FIG. 2

PLATE 2

FIG. 1. Extinction of a shallow lake. The disappearance of the water is so recent that some aquatic vegetation has persisted. Part of Crosby Lake, Oakland County, Michigan

FIG. 2. Soil profile in an old lake bed eight miles west of Seney, Schoolcraft County, Michigan



FIG. 1



FIG. 2

ABSORPTION OF SELENIUM AND ARSENIC BY PLANTS FROM SOILS UNDER NATURAL CONDITIONS¹

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It has recently been demonstrated that arsenic in the form of sodium arsenite is effective in counteracting the toxicity of selenium (3, 4), and in view of this fact the arsenic content of seleniferous feeds is being studied. Consequently, it has been advisable to study the arsenic content of soils and of their parent geologic formations and also the availability to plants of arsenic in soils.

The arsenic content of several samples from geologic formations of South Dakota has been found, in general, to be higher than their selenium content.² Williams and Whetstone (5) have reported the arsenic content of several soil and plant samples from various parts of the United States and Mexico. The values for soils ranged from 0.3 to 40.0 p.p.m. of arsenic and those for vegetation, unless collected from soils previously treated with arsenic, were in only a few cases higher than 3.0 p.p.m.

This study was made to determine what natural relationship, if any, exists between arsenic and selenium in soils. Several samples of naturally seleniferous soils and of plants which had grown in these soils, all of which had been analyzed for selenium, were available for the study.

DESCRIPTION OF SAMPLES

In connection with grazing experiments being conducted in a seleniferous area by the South Dakota Experiment Station, 10 plots, each 2 rods square, have been kept fenced for about 2 years. These plots have been used for studies on selenium in plants and soils. During the summer of 1939, soil samples were collected from each plot at three different depths: 0-12, 12-24, and 24-36 inches. Several plant samples, also, were taken at various times during the summer. After selenium determinations had been made on these samples, they were available for arsenic analysis.

METHODS OF ANALYSIS

Selenium was determined by the digestion distillation method (2); arsenic, by the method of Klein and Vorhes (1). Both selenium and arsenic are

¹ Approved for publication by the director of the South Dakota Agricultural Experiment Station as Journal Series No. 131.

² Unpublished data.

reported as parts per million of the element, based on the air-dry weight of the sample.

RESULTS AND DISCUSSION

The results for the analyses of the soils for selenium and arsenic are given in table 1.

No definite correlation between the selenium and arsenic contents of the soils studied is apparent. In all cases the arsenic content, which ranged from 7.1 to 18.4 p.p.m., was greater than the selenium content, which ranged from 1.15 to 5.0 p.p.m. With few exceptions, the plants which had grown in

TABLE 1
Selenium and arsenic contents of soil samples

PLOT NUMBER	DEPTH	Se CONTENT	As CONTENT	PLOT NUMBER	DEPTH	Se CONTENT	As CONTENT
	<i>inches</i>	<i>p.p.m.</i>	<i>p.p.m.</i>		<i>inches</i>	<i>p.p.m.</i>	<i>p.p.m.</i>
1	0-12	1.5	9.3	6	0-12	1.15	9.2
	12-24	3.68	8.8		12-24	1.65	9.6
	24-36	5.0	9.0		24-36	1.4	8.8
2	0-12	3.9	17.0	7	0-12	2.0	9.6
	12-24	3.5	13.0		12-24	1.75	9.5
	24-36	3.9	16.0		24-36	1.65	18.4
3	0-12	1.55	9.1	8	0-12	4.0	9.6
	12-24	1.5	9.3		12-24	4.5	9.8
	24-36	2.4	8.5		24-36	5.0	9.2
4	0-12	3.75	12.6	9	0-12	1.4	10.1
	12-24	3.65	7.7		12-24	2.4	16.4
	24-36	4.9	9.8		24-36	2.75	9.8
5	0-12	2.05	8.6	10	0-12	2.4	8.6
	12-24	3.8	8.5		12-24	3.15	15.6
	24-36	3.25	7.1		24-36	3.75	9.1

these soils, however, were found to contain more selenium than arsenic, as is indicated in table 2.

The arsenic in the soils studied is much less available to plants than is the selenium. None of the plants contained more than 4.3 p.p.m. of arsenic, whereas 266.7 p.p.m. of selenium was found in one sample. The variation in the selenium content of the plants is no doubt due largely to differences in the abilities of the several species of plants to absorb the element and to variations in the availability of the element. Apparently none of the species of plants analyzed are capable of absorbing from soils large quantities of arsenic unless this is in an easily available form.

Experiments are under way to determine whether the arsenic occurring

TABLE 2
Selenium and arsenic contents of plant samples

LABORATORY NUMBER	PLOT NUMBER	COMMON NAME	SCIENTIFIC NAME	STAGE OF GROWTH	Se CON- TENT	As CON- TENT
					p.p.m.	p.p.m.
111	1	Sunflower	<i>Helianthus</i> sp.?	Blossom	46.6	3.3
112	1	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Early heading stage	18.2	2.3
113	1	Feather grass	<i>Stipa viridula</i> Trin.	Seed shedding	12.8	2.3
116	2	Feather grass	<i>Stipa viridula</i> Trin.	Seed shedding	4.0	2.0
117	2	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Preheaded	0	1.2
118	2	Blue grama grass	<i>Bouteloua gracilis</i> (H.B.H.) Lag.	Fertilized	1.0	1.7
119	3	Sunflower	<i>Helianthus</i> sp.?	Late blossom	4.3	1.8
120	3	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Preheaded	3.2	3.5
122	3	Blue grama	<i>Bouteloua gracilis</i> (H.B.H.) Lag.	Fertilized	4.4	1.7
123	3	Gum plant	<i>Grindelia squarrosa</i> (Pursh) Dunal.	Early bud	30.0	3.2
126	4	Needle grass	<i>Stipa comata</i> Trin. & Rupt.	Seed shed	4.0	3.0
128	4	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Preheaded	4.8	4.3
130	5	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Preheaded	5.4	2.3
132	6	Lead plant	<i>Amorpha</i> sp.?		0	3.2
133	6	Side oat grama	<i>Bouteloua curtipendula</i> (Michx.) Torr.	Fertilized	1.0	4.3
135	7	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Preheaded	4.4	1.7
137	7	Blue grama grass	<i>Bouteloua gracilis</i> (H.B.H.) Lag.	Late fertilized	3.3	2.7
139	8	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Preheaded	7.4	2.2
142	8	Gum plant	<i>Grindelia squarrosa</i> (Pursh) Dunal.	Early bud	266.7	1.7
143	9	Feather grass	<i>Stipa viridula</i> Trin.	Seed shed	11.0	1.8
145	10	Sunflower	<i>Helianthus</i> sp.?	Late blossom	7.0	4.2
146	10	Western wheat grass	<i>Agropyron smithii</i> Rydb.	Preheaded	6.0	2.8
149	10	Feather grass	<i>Stipa viridula</i> Trin.	Seed shedding	4.0	3.0

naturally in plants is effective in preventing the toxic action of selenium in animals. If this naturally occurring arsenic proves to be antagonistic to selenium toxicity in animals, it will be another factor to consider in connection with studies on the toxicity of seleniferous vegetation.

SUMMARY

Several soil and plant samples have been analyzed for selenium and arsenic.

No correlation between the selenium and arsenic content of soils is apparent from this study.

The arsenic in the soils studied has been found to be considerably less available to plants than is the selenium.

REFERENCES

- (1) KLEIN, A. K., AND VORHES, F. A. 1939 Determination of arsenic. *Jour. Assoc. Off. Agr. Chem.* 22: 121-130.
- (2) MOXON, A. L. 1937 Alkali disease or selenium poisoning. S. Dak. Agr. Exp. Sta. Bul. 311.
- (3) MOXON, A. L. 1938 The effect of arsenic on the toxicity of seleniferous grains. *Science* 88: 81.
- (4) MOXON, A. L., AND DuBois, K. P. 1939 The influence of arsenic and certain other elements on the toxicity of seleniferous grains. *Jour. Nutrition* 18: 447-457.
- (5) WILLIAMS, K. T., AND WHETSTONE, R. R. 1940 Arsenic distribution in soils and its presence in certain plants. U. S. Dept. Agr. Tech. Bul. 732.

THE NATURE OF THE CATALYST CAUSING THE HYDROLYSIS OF UREA IN SOILS

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Urea may be hydrolyzed to ammonium carbonate in soils by microorganisms, as is generally recognized, and by thermolabile catalysis, as has been recently demonstrated (1, 3). In the one soil reported on, nearly as much urea was hydrolyzed in the presence of toluene as in its absence. Later tests with a greater number of soils showed, in general, the same type of behavior. It is too early to generalize, but if the type of behavior so far found is widespread, catalysis independent of direct microbial activity will be found to be the principal agency responsible for the hydrolysis of urea in most natural soils.

The data previously published indicate that the catalyst or catalysts are for the most part thermolabile, as the catalytic activity virtually disappeared when the moistened soil was preheated to about 85°C. Some soils, at least, preheated in this way still show a small amount of catalytic activity, hence a catalyst or catalysts that are thermostable exist in soils.

A thermolabile catalyst in a biological system strongly suggests an enzyme. Other data collected support such a view. After the adsorptive capacity² of the untreated soil was satisfied, a constant rate of percolation at a constant temperature, even with widely varying concentrations of urea, gave almost constant reductions in the urea concentration of the percolating solution. This type of behavior; namely, a constant catalytic rate independent of the concentration of the substrate, has been found with certain enzymes, including urease (4, 16). Aqueous extracts of soils showed no catalytic activity. The best initial hypothesis to explain these, as well as other data reported, envisioned the enzyme, urease, adsorbed on the soil colloids but with its reactive group or groups still free to bring about chemical change. This paper presents data arising from a more critical examination of such a hypothesis.

ATTEMPTED ELUTION

Before urease, as well as some other enzymes, were available in a crystalline form, partial purification and concentration were generally effected by ad-

¹ The analyses reported herein were kindly carried out by J. M. Weiler and J. A. Garibaldi, technicians in the Division of Agronomy, Davis.

² In a previous study (1) the marked reductions in concentrations of the first percolates were attributed mainly to adsorption. As was to be expected, adsorption with each successive percolate rapidly diminished as a factor in removing urea from solution.

sorbing the enzyme on a specific colloid, separating the colloid from the liquid phase, and then washing out the enzyme with a solution of an eluent, a specific substance which would displace the enzyme from the colloid. Though this method of adsorption is not used in the purification and concentration of urease so generally as before, still urease has been adsorbed on kaolin (19), on $\text{Ca}_3(\text{PO}_4)_2$ (10, p. 349), on alumina (19), on fibrin flakes (8), and on cholesterol (9); and from some of these it has been eluted by such substances as soda and phosphate buffers. Some of these colloids occur in soils; probably others do not.

Attempts to elute urease activity from our soils by means of various solutions, including 0.1 *N* H_2SO_4 , H_3PO_4 , K_2HPO_4 , and acetic acid, were unsuccessful. Each solution (150 cc.) was mixed with 100 gm. of soil and filtered. A 50-cc. aliquot of the filtrate and about 1 cc. of toluene were added to 20 cc. of urea solution containing 50 millimols per liter and incubated at 40°C. for 16 hours. No ammonia was in evidence with Nessler's reagent after the incubation.

In the previous study (1) the catalytic activity of the soil declined slightly as the percolation of the urea solutions was extended. This could be attributed to the enzyme's combining with its substrate and being eluted from the soil. In consequence, later tests included 0.5 and 1.5 *M* urea among the eluents tested. Neither of these solutions was able to elute more than traces of activity even when the ensuing incubation periods were extended to several days. This was true also of 1 and 3 *N* NaOH and NH_4OH . If the small amounts of $(\text{NH}_4)_2\text{CO}_3$ frequently found in these incubation tests with the filtrates and percolates indicate catalytic activity eluted from the soil and not experimental error, the activities thus eluted represent only very small fractions of the activities shown by the soils themselves. Further experiments with other combinations of soils and eluting solutions may possibly demonstrate activities which can be separated from soils. Subrahmanyam (14) reported the elution with glycerine of a deaminase from water-logged soils. When a technic similar to that described above was used, 0.5 *M* glycerine solutions were able to elute only traces of activity from our soils.

ANTISEPTICS AND INACTIVATORS

Though efforts to elute any appreciable activity from the soil were unsuccessful, other criteria are available to test for the presence of urease adsorbed on the soil colloids; namely, the reactions of this hydrolytic activity in soils to antiseptics and inactivators whose reactions on urease are known.

Fifty per cent alcohol is believed by the writer to be concentrated enough to act as a very effective though perhaps not an absolutely complete inhibitor of microorganisms. Solutions of urea in this solvent were percolated through 400-gm. lots of soil in glass percolators by adding to each percolator a 140-cc. portion to wet the soil initially and successively 75-cc. portions at 12-hour intervals thereafter. The percolations, unless otherwise specified, were

carried on in a room maintained at a constant temperature of about 30°C. Residual urea in the various percolates was determined by the Van Slyke and Cullen method essentially as described by Hawk and Bergeim (7, p. 709). Figure 1 shows that considerable catalysis (the difference between the activities in normal and preheated soil) took place in Yolo fine sandy loam with the 50 per cent alcoholic solutions. It is true that no constant rate of reduction in concentration of urea was observed. It may be that the alcohol had a gradually injurious effect on the catalyst.

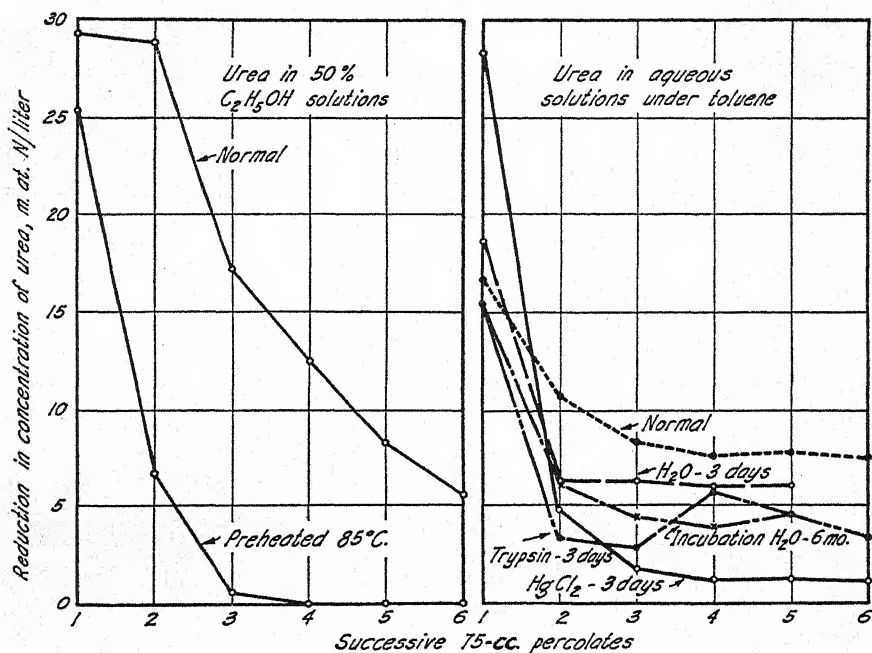


FIG. 1. REDUCTION IN CONCENTRATION OF UREA SOLUTIONS PERCOLATED THROUGH 400 GM. OF YOLO FINE SANDY LOAM AS AFFECTED BY VARIOUS PRETREATMENTS AND ANTISEPTICS

The successive additions of the urea solutions containing 39.8 m. at.³ N per liter were at 12-hour intervals.

In figure 2, comparable studies with Nord loam show less deleterious effect of the alcohol on the activity of the catalyst. Preheating the Nord loam caused the catalytic effect to disappear entirely, since with the fourth successive percolate the urea solution came through virtually unchanged. These data are evidence again that the hydrolysis of urea in soil is catalytic and that the catalyst is destroyed or inactivated by preheating the moist soil to 85°C.

Traces of $HgCl_2$ are known to inhibit the activity of urease, and the proteolytic enzyme, trypsin, has been shown to inactivate urease by digestion in a

³ As used in this paper, m. at. indicates milligram atoms.

short time (6, 15). Lots of soil, therefore, were pretreated respectively with HgCl_2 and trypsin, moistened with water, and allowed to incubate with occasional mixing for varying lengths of time. Finally each lot of soil was spread on the laboratory table to dry. These lots of soil were then percolated as before with solutions of urea, and the residual urea was determined in the percolates. To the percolates from the HgCl_2 -treated soil, KCN in excess was added to eliminate the inhibiting effect of any HgCl_2 which might have been leached out of the soil. It will be observed in figure 1 that both the

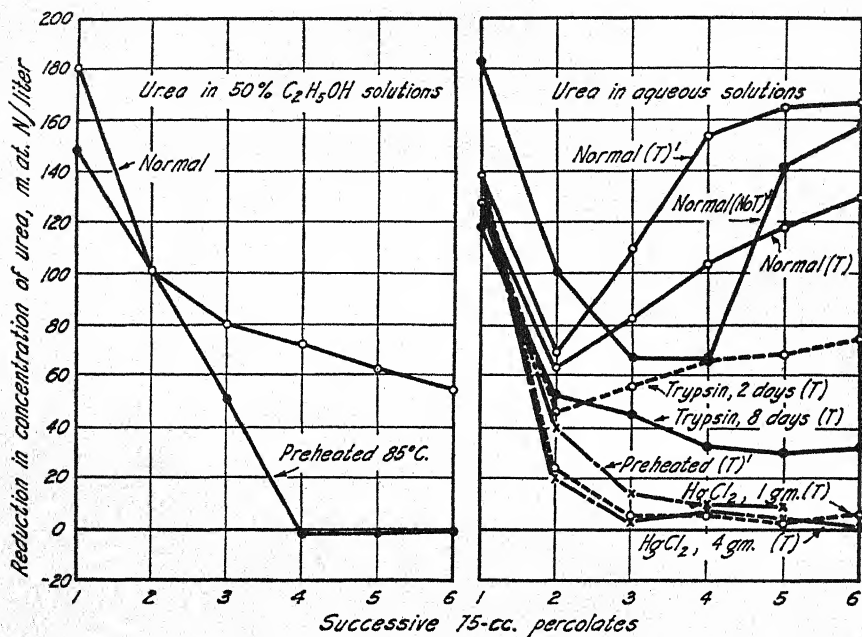


FIG. 2. REDUCTION IN CONCENTRATION OF UREA SOLUTIONS PERCOLATED THROUGH 400 Gm. OF NORD LOAM AS AFFECTED BY VARIOUS PRETREATMENTS AND ANTISEPTICS

T-under toluene,¹ in experiment 1, solutions contained 194.4 m. at. N per liter as urea; in the other, 193.5. As in figure 1, successive additions were at 12-hour intervals.

HgCl_2 and the trypsin pretreatments reduced the catalytic activity in the Yolo soil.

More extensive trials were made with Nord loam because of the much greater activity which it had shown in previous tests. As disclosed in figure 2, predigestion with trypsin (1 gm. per kilogram of soil) for 2 days caused a material reduction in the activity; predigestion for 8 days reduced the activity still further. The pretreatment with HgCl_2 , whether 1 gm. or 4 gm. per kilogram of soil, for 2 days reduced the activity almost to zero; in fact, the activity of the HgCl_2 -treated soil was less than that of the preheated soil. The preheated Nord loam has shown a small but appreciable thermostable catalytic activity toward urea in aqueous solutions.

Additional tests with various pretreatments were conducted with three other soils, the results of which are reported in table 1. Here it will be observed that with Fresno fine sandy loam almost identical results were obtained with percolation in the presence of and in the absence of toluene. Preheating the Fresno soil removed this catalytic effect. With Aiken loam, differences of some magnitude occurred among the first percolates. It may be that dissimilarities

TABLE 1

Concentration of urea in successive 75-cc. percolates from 400-gm. portions of soils as influenced by different antiseptics and various pretreatments

Results in milligram atoms N per liter

SOIL AND PRETREATMENT	ANTISEPTIC*	CONCENTRATION OF UREA IN						
		Original solution	Successive percolates					
			1	2	3	4	5	6
Fresno fine sandy loam								
None.....	C ₆ H ₅ CH ₃	38	35	33	34	34	34	37
None.....	None	38	34	30	34	36	36	35
Preheated 85°C.....	C ₆ H ₅ CH ₃	48	23	48	48	48		
Aiken loam								
None.....	None	38	7	13	10	11	12	13
None.....	C ₆ H ₅ CH ₃	38	24	15	10	11	11	11
Preheated 85°C.....	C ₆ H ₅ CH ₃	38	22	31	34	35	36	36
Autoclaved 120°C.....	C ₆ H ₅ CH ₃	38	32	31	32	33	34	35
Predigested trypsin†.....	C ₆ H ₅ CH ₃	38	13	24	25	25	26	27
Yolo silt loam								
None.....	C ₆ H ₅ CH ₃	195	108	151	162	175	162	161
Preheated 85°C.....	C ₆ H ₅ CH ₃	195	87	163	186	195	190	197
Predigested trypsin†.....	C ₆ H ₅ CH ₃	195	95	166	176	181	170	173
Yolo fine sandy loam								
None.....	C ₆ H ₅ CH ₃ ‡	40	24	33	34	36	36	36
None.....	CHCl ₃ ‡	40	24	32	34	34	34	33
None.....	CS ₂ ‡	40	24	33	34	35	35	35

* Present during percolation, at constant temperature, unless otherwise noted, of 30°C.

† One kilogram soil plus 1 gm. trypsin, mixed, moistened, incubated 3 days, and then dried at room temperature.

‡ One per cent of the percolating solution. Percolations at constant temperature of slightly less than 20°C.

in the state of aggregation of the samples or in the way in which the solutions or the antiseptics allowed the urea solution to come into contact with the soil caused these differences. Beginning with the third percolate there was, however, an almost constant reduction in the concentration of the urea. Only a small difference in activity occurred between lots of the same soil percolated under toluene and without it. Aiken soil predigested with trypsin showed

approximately one-half the catalytic activity of the normal soil. Preheating of the Aiken soil to 85° and to 120°C. (the latter in the autoclave) resulted in almost identical data, but these strenuous treatments did not entirely remove all the catalytic activity. Some thermostable catalysis remains. Predigestion of Yolo silt loam with trypsin reduced the catalytic activity somewhat, but not, perhaps, to the same degree as in the other soils. Preheating of this soil to 85°C. resulted in the complete disappearance of this activity.

Just before this manuscript was prepared, the effects of other antiseptics were tested. The same technic was used as before except that the percolations were carried on in a laboratory at a temperature of slightly less than 20°C. The results with Yolo fine sandy loam are given at the bottom of table 1. Though the various antiseptics produced results differing slightly in absolute amounts, each gave evidence of catalysis, and all showed the same

TABLE 2
Effect of various urease-inhibitors upon the hydrolysis of urea in two soils
Nitrogen in milligram atoms per kilogram soil

SOIL AND PRETREATMENT	NITROGEN ADDED AS UREA	NITROGEN RECOVERED AFTER 7 DAYS' INCUBATION		
		As urea	As ammonia	Total
Nord loam C-18				
None.....	67.0	66.3	66.3
Preheated 85°C.....	67.0	55.7	9.9	65.6
Predigested with hydroquinone.....	67.0	63.4	1.7	65.1
Predigested with catechol.....	67.0	58.6	6.1	64.7
Yolo fine sandy loam C-68				
None.....	13.3	1.1	12.2	13.3
Predigested with hydroquinone.....	13.3	11.3	0.5	11.8
Predigested with catechol.....	13.3	13.1	0.8	13.9

type of behavior. Unreported tests with Nord loam showed marked catalysis with each antiseptic, but some differences occurred among the results with the different antiseptics. These will need further study.

Another type of urease inhibitor requires attention. A small amount of either hydroquinone or catechol is known (12) to inhibit almost completely the activity of urease. In consequence, kilogram lots of two soils, as reported in table 2, were respectively moistened with about 350 cc. of 0.5 *M* hydroquinone and 0.2 *M* catechol, incubated under toluene for 3 days at 30°C., and then dried at room temperature.

In these tests 75-gm. lots, pretreated as indicated in table 2, were added to incubation jars, urea solution was added, and incubation was continued for 7 days under toluene. Extracts of these lots of soil with *N* KCl at pH 1, essentially the Olsen method as described by Wright (20), were made, and ammonia was determined by aeration. The inhibiting effect of either the

hydroquinone or catechol cannot be removed, as was that of HgCl_2 , by the addition of KCN. Hence the residual urea in the percolates was determined by the gravimetric xanthidrol method of Fosse (11, p. 553). The data show that in the normal soil under toluene, Nord loam completely changed the urea to ammonium carbonate. In the preheated soil about one-sixth of the urea was so changed. The pretreatment with hydroquinone and catechol resulted in the transformation of still less of the urea. Again in the normal Yolo soil nearly all the urea was transformed to ammonia. The pretreatments with hydroquinone and catechol, however, prevented all but the slightest transformation.

ADSORPTION OF UREASE BY SOIL

Failures to elute catalytic activity from the soil neither strengthen the initial hypothesis that the activity is caused by urease adsorbed on the soil solids nor constitute critical evidence against it. There may be so many different kinds of colloids in the soil that a specific eluent might release the enzyme from one colloid but would bring it into the proper range of conditions for its adsorption by another. The ability of the soil to adsorb urease is, therefore, worthy of investigation. Since the preheated Yolo fine sandy loam has almost no catalytic activity for urea, it was considered to be a good medium for the study of the adsorption of urease by the soil.

Equilibrium studies

Equilibrium studies were carried on between the preheated soil and urease solutions of various concentrations. In general, 50 gm. of preheated soil were shaken with 100 cc. of the urease solution being tested. After a few shakings the suspension was allowed to settle for 2 hours and was then filtered. A 50-cc. aliquot of the filtrate was added to 20 cc. of a solution containing 50 millimols of urea per liter. At the same time, 50 cc. of standard urease solutions were added to similar 20-cc. aliquots of the standard urea solution mentioned. After incubation at 38°C . for 3 hours, ammonia was determined by aeration. The computed concentrations of urease per liter in equilibrium with the calculated amounts of urease adsorbed per kilogram of preheated soil are given in figure 3. A curve is drawn fairly close to most of the experimental points. It will be observed that all of the activity, equivalent to 0.5 gm. of urease per kilogram of soil, disappeared from solution, presumably being adsorbed by the soil. This amount is higher than the natural catalytic activity found in the normal Yolo fine sandy loam.

Reversibility of urease adsorption

In another experiment the reversibility of the adsorption of added urease was investigated. A 300-cc. portion of the urease solution being tested was added to 200 gm. of the preheated Yolo soil. After 12 hours the excess solution was allowed to drain away through the percolator. At 12-hour intervals

thereafter, the soil was successively leached with 150-cc. portions of distilled water, and the urease activity of the successive leachates was determined by adding aliquots to standard urea solutions and titrating, as in the Marshall method (7, p. 712), the ammonia formed after incubation for 12 hours at 30°C. The activity found was standardized against known solutions of urease handled in a comparable way. The results of this study are given in table 3.

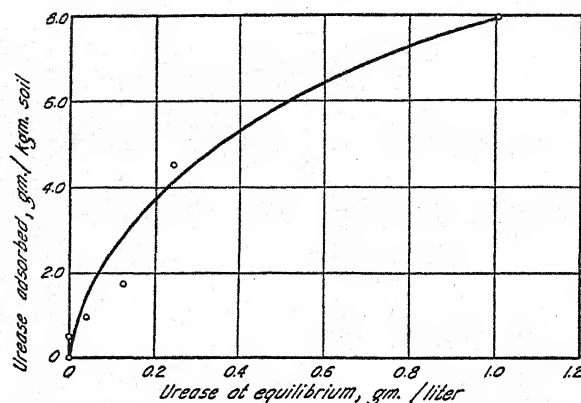


FIG. 3. ADSORPTION OF THE ACTIVITY OF JACK-BEAN UREASE BY PREHEATED YOLO FINE SANDY LOAM—EQUILIBRIUM STUDIES

TABLE 3

Retention of the activity of Jack-bean urease by preheated Yolo fine sandy loam and recovery of some of the activity by subsequent leaching with water

UREASE ADDED IN 300 CC. TO 200 GM. SOIL.....mgm.	15	150	300	600
Urease recovered from:				
200 cc. urease filtrate.....mgm.	0.0	1.4	1.4	1.4
1st 150 cc. H ₂ O leachate.....mgm.	0.0	2.0	3.0	5.6
2nd 150 cc. H ₂ O leachate.....mgm.	0.0	2.3	2.7	4.7
3rd 150 cc. H ₂ O leachate.....mgm.	0.0	1.5	2.3	4.4
4th 150 cc. H ₂ O leachate.....mgm.	0.0	1.1	1.5	3.0
Total recovered.....mgm.	0.0	8.3	10.9	19.1

Here it will be observed that 15 mgm. of urease have been completely adsorbed and retained against leaching by 200 gm. of soil. This would be equivalent to 75 mgm. of urease per kilogram of soil. Higher concentrations of urease showed incomplete adsorption by the soil. The activity leached out, however, was always less than 5 per cent of the amount added. It should be observed that in the case of the highest amount of urease completely retained, the activity was slightly greater than that found in the normal soil.

Percolation studies

A method has been described (2) for determining by plant response the retention by soils of nutrient ions and compounds from percolating solutions. The method was modified to study the adsorption by the soil of urease from solution. Different columns of pots were percolated with just enough urease solution to wet, with slight excess, the dry preheated soil in the pots. After percolation had ceased, the columns were dismantled and the soil in each pot was dried out at temperatures not over 40°C. This procedure usually required 48 to 72 hours. The lots of soil were then returned to the respective pots, and the catalytic activity of the soil was determined. The results of these tests are shown in table 4. Columns percolated with solutions containing 5 mgm. of urease per liter are suggestive of some retention of activity in the top pot. Columns percolated with 50 mgm. of urease per liter showed definite evidence of retention of activity in the top pot; in fact, the activity was of about the same order of magnitude as that of normal soil. There is a slight suggestion that some of the urease activity might have been carried into the second pot of the column. The decline of the added activity with successive percolates of urea solution was rapid. Somewhat lower values were obtained in experiment 2 for 50 mgm. of urease per liter, undoubtedly because of some chance difference in procedure. With the solution of 500 mgm. urease per liter the retention in the top pot was marked.

NATURAL VS. ADDED ACTIVITY

Evidence so far collected suggested that the soil catalyst has many of the characteristics of the laboratory enzyme, Jack-bean urease. On the other hand, certain behavior of the added laboratory urease suggested that it was not so resistant to decomposition in the soil as was the natural catalyst. That laboratory urease added to preheated soil rapidly loses its activity, is shown by the data in tables 3 and 4, especially when compared with the data in table 5. Here, dry urease was added to 400 gm. dry preheated soil and then percolated with various concentrations of urea without any intervening period of incubation. The 50 mgm. of urease per kilogram of soil removed almost all the urea from solution, especially after the first two percolates were collected. Many of the pretreatments of the soil with urease solution, as reported in tables 3 and 4, resulted in the addition of even greater amounts of urease in solution; thus in table 4 the top pot of the 500-mgm. treatment would have received at least 80 mgm. of urease even if none of it were retained. If all the urease were retained by the soil of the top pot, then this pot would have received 250 mgm. of urease, ten times as much as that received in a dry condition by the soil in table 5. It will be observed too in table 4, that the added activity of the urease rapidly disappeared with successive percolates, indicating that the added urease was not nearly so stable as the natural activity of the soil.

TABLE 4

Retention of the activity of Jack-bean urease by preheated Yolo fine sandy loam in columns of pots as shown by subsequent percolation trials with urea solutions

Results in milligram atoms N per liter

CONCENTRATION OF PERCOLATING UREASE SOLUTION	ORDER OF POTS IN PERCOLATING COLUMN	CONCENTRATION OF UREA IN						
		Original solution	Successive 75 cc. percolates					
			1	2	3	4	5	6
mgm./l.								

Experiment 1								
5	Top 1	20.0	8.2	15.4	17.2	17.4		
	Middle 2	20.0	12.0	18.6	17.8	17.8		
	Bottom 3	20.0	10.8	16.6	18.6	20.0		
50	Top 1	20.0	4.8	5.2	11.0	15.0		
	Middle 2	20.0	8.2	19.4	18.0	18.2		
	Bottom 3	20.0	8.0	18.0	18.6	19.0		

Experiment 2								
50	Top 1	38.9	20.8	35.9	37.9	38.2	38.1	38.6
	Middle 2	38.9	26.4	36.8	38.0	38.0	38.5	38.8
	Bottom 3	38.9	26.1	37.1	37.3	39.4	39.3	39.4
500	Top 1	38.9	3.7	4.8	8.6	14.2	18.7	23.9
	Middle 2	38.9	27.2	36.4	37.4	37.4	37.6	37.6
	Bottom 3	38.9	26.1	36.4	37.0	39.7	39.7	39.8

TABLE 5

Effect of dry Jack-bean urease added to Yolo fine sandy loam preheated to 105°C. upon the concentration of urea subsequently percolated through it

Results in milligram atoms N per liter

UREASE ADDED mgm./kgm.	CONCENTRATION OF UREA IN				
	Original solution	Successive 75-cc. percolates			
		1	2	3	4
0	0.0	2.6	0.8	0.0	0.0
	26.1	20.4	25.6	24.8	25.6
	51.6	35.2	51.0	50.0	51.8
	104.8	61.2	101.4	97.0	104.8
50	0.0	3.8	0.0	0.0	0.6
	26.1	6.8	1.6	0.6	0.8
	51.6	15.6	2.0	0.6	0.2
	104.8	30.2	3.4	0.6	1.0

It seemed desirable, therefore, to conduct simultaneous percolations and incubations with soils variously treated. These data are shown in figure 4. The lots of Yolo fine sandy loam used were (a) normal soil, (b) soil preheated to 85°C., (c) similarly preheated soil plus 50 mgm. dry Jack-bean urease per kilogram of soil, (d) preheated soil plus 50 mgm. urease per kilogram of soil as in (c) but moistened, incubated for 7 days, and then redried at room temperature. The activity of this incubated soil was but little, if any, higher than that of the preheated soil. The soil not incubated showed high initial

TABLE 6

Effect of various pretreatments of Yolo fine sandy loam upon ammonia formed from urea during incubation under toluene*

Results in milligram atoms of N per kilogram of soil

SOIL PRETREATMENT	NITROGEN TRANSFORMED TO NH_3 DURING INCUBATION	
	1 day	3 days
None.....	2.25	5.86
Preheated.....	0.23	0.45
Preheated plus 50 mgm. urease/kgm.		
Not preincubated.....	26.7	17.7
Preincubated 7 days, dried.....	1.27	1.45

* Urea added at the rate of 26.7 m. at. N per kilogram of soil.

TABLE 7

Effect of urease decomposition upon the average yield of triplicate pot cultures of Darlo plants

	NO SOIL PRETREATMENT	SOIL PREHEATED TO 85°C.		
			Urea 50	Urease 100
Material added.....	0	0		
Amount per pot.....mgm.	0	0		
Yield per pot				
Green.....gm.	1.18	2.23	6.49	3.35
Dry.....gm.	0.25	0.44	1.27	0.68

activity, but by the time the sixth percolate was collected this activity had rapidly diminished. If the percolations had been continued long enough, it is possible that this activity might have disappeared altogether. The activity of the normal soil, however, after the third percolate, was virtually constant.

Additional trials were made by incubation in which 75 gm. of soil similarly treated were incubated with urea under toluene. At the end of the incubation periods the amount of ammonia formed was extracted by the Olsen method (20, p. 127), determined by aeration, and reported in table 6. Again the normal soil transformed appreciable amounts of urea to ammonia. The pre-

heated soil showed little transformation. The preheated soil to which urease was added, but not preincubated, showed complete transformation of the urea to ammonia. The urease-treated soil, preincubated for 7 days, showed even less activity than the normal soil but slightly more than the preheated soil. These data show that the natural activity of the soil was more resistant to destruction than was the laboratory urease added to preheated soil.

Data collected in connection with another phase of the problem were also suggestive of the decomposition of the added Jack-bean urease in the soil. Certain amendments, including urea, and urease were added to preheated soil in triplicate cultures in pots cropped to milo. The yields are reported in table 7. Differences between any two comparable averages are statistically significant (5, p. 114), P being less than 0.01 in each case. Urea gave a marked response in yield. We may infer from these data that the soil under these experimental conditions was deficient in nitrogen and that the increased growth from the addition of the urease was due to the partial breakdown of this proteinaceous material to give available forms of nitrogen.

DISCUSSION

Evidence presented in a previous paper (1) showed that the hydrolysis of urea in the soil reported on was due largely to a thermolabile catalyst or catalysts rather than to microorganisms. Later tests have shown the same type of behavior on many more soils. In extending this type of evidence, hydrolysis has been demonstrated by the technic of percolating through soils urea in 50 per cent alcoholic solution and in aqueous solutions under the three antiseptics: toluene, chloroform, and carbon disulfide. These data were in line with the previously postulated catalytic nature of the process.

In 1877 Schloesing and Müntz, as cited by Russell (13, p. 24), investigated the purification of sewage percolated slowly through a filter of sand and limestone. After an initial period in which ammonia was the chief form of nitrogen in the percolate, nitrates began to appear and to increase until no more ammonia came through. A little chloroform vapor stopped the transformation of ammonia to nitrate. Soon Warrington showed that this same transformation in soils could also be stopped by chloroform and carbon disulfide. Later the bacterial nature of the process was completely demonstrated. If then, the stoppage by antiseptics of the transformation of ammonia to nitrate was evidence of the activities of the microorganisms, the lack of any effect by antiseptics on a transformation is good evidence that that transformation is not directly microbial, but chemical.

A former study (1) suggested that the thermolabile catalyst in soils responsible for the hydrolysis of urea might be urease adsorbed on the soil colloids. Evidence for such a hypothesis presented in this paper is the adsorption of the activity of Jack-bean urease by soils. On the other hand, neither the native activity nor the activity added in Jack-bean urease has been successfully eluted from the soil. Similarities of behavior of the native activity to that

of laboratory urease in addition to the effects of heat and antiseptics are as follows: HgCl_2 inactivated the activity of the native soil, as well as that of the laboratory urease; hydroquinone and catechol did likewise. Similarly, trypsin decomposed laboratory urease, thereby destroying its power of hydrolysis. Treating soils with trypsin materially reduced the catalytic activity. These markedly different types of behavior, all of which individually are characteristics of urease, indicate strongly the enzymatic nature of the catalytic activity. A more direct quantitative comparison of the rates of inhibition of the activities of the natural soil and of added laboratory urease with given quantities of inhibitors and proteolytic enzymes are likely to be fruitless because of the possible dissipating action of soil.

As the studies progressed, indications appeared that the activity of the added urease was not stable. A special study was therefore made to test

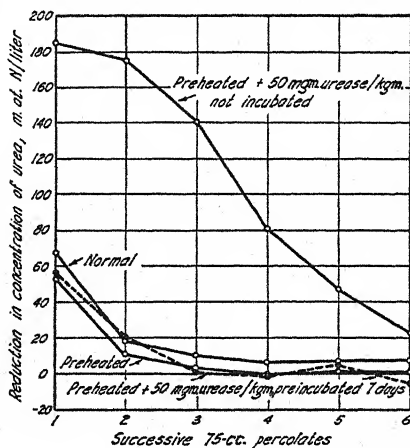


FIG. 4. COMPARATIVE STABILITY OF THE CATALYTIC ACTIVITY OF NORMAL YOLO FINE SANDY LOAM AND OF THAT ADDED IN JACK-BEAN UREASE TO PREHEATED SOIL

this factor. The results, reported in figure 4 and in tables 6 and 7, showed that incubation rapidly reduced or destroyed the activity of the added urease, and that while the subsequent percolation tests were being conducted the activity of the added urease rapidly diminished, whereas the native activity had only a slight tendency to diminish. This fact throws some doubt on the absolute values reported in the early tables where urease was added to a soil. From a comparative standpoint, they all retain qualitative values. At this time many new phases of the subject seem to require immediate consideration; hence, repeating these studies to improve their absolute values seems of questionable concern.

Data in figures 1 and 2 and in tables 1 and 2 indicate that the soil catalyst and the laboratory enzyme, urease, have many properties in common. If the soil catalyst is an enzyme, data in figure 4 and in tables 6 and 7 indicate

that it is more resistant to decomposition in soils than is added urease. It may be an enzyme protected from decomposition by some resistive combination with inorganic and organic substances in the soil. Waksman and Iyer (17) have synthesized some lignoproteinates the properties of which were similar to material separated from soil. The synthetic materials resembled the natural substances in resistance to attack by microorganisms. Later (18) they concluded from further studies:

Proteins are capable of forming with lignins... complexes which render them more resistant to attack by microorganisms. . . . There is no doubt that the formation of a ligno-protein is not an instantaneous process.

Conceivably, urease might be one of the proteins capable of forming with the lignins complexes which would be resistant to decomposition and still leave its reacting group or groups free to perform the enzymatic function. The data in this paper are in the main in agreement with such a concept, except that no catalytic activity was separated from the soil by technics (i.e., extraction with NaOH, NH_4OH , etc.) which are known to extract some ligno-proteins.

It is too early to indicate what the exact protective action in this case would be, but conceivably there might be some protective combination which would prevent the decomposition of the urease in this union. The proteolytic enzyme, trypsin, however, in a few days' digestion was able materially to reduce the amount of activity in these soils. Presumably, therefore, decomposition might eventually break down even this protected catalyst. From the data contained herein and in previous papers, it seems probable that an enzyme, similar in many respects to urease, occurs in many soils and that this is the chief agency in converting urea to ammonium carbonate in soils. More investigations should be made on a greater variety of soils to test the presence or absence of this enzymatic activity.

To the agronomist, however, the questions of the role of the catalytic activity and the possible effect that it may have upon plant growth are of interest. Crop growth may be determined by the rate of supply of the essential element most deficient. The presence of catalysts or enzymes such as urease may have an important role in determining the speed with which the lacking element, say nitrogen, is supplied to the plant. The study of possible catalytic activities in soils would seem, then, to offer a fruitful field of research from a purely scientific standpoint as well as from an agronomic and utilitarian viewpoint. The process of transforming urea to ammonium carbonate seems to be chiefly catalytic and presumably enzymatic. The transformation of ammonia to nitrate appears to be almost entirely bacteriological. With this new concept of these transformations in the soil, it would seem desirable to recheck all the transformations formerly attributed to microorganisms to determine which ones are entirely due to them and which ones can be accom-

plished catalytically.⁴ In the step-by-step breakdown of soil organic matter to furnish nitrates in the soil, it may be that some steps are entirely enzymatic and some entirely microbiological. Hence, the rate of decomposition and consequently the rate of supply of some of the nutrients may be dependent upon the proper functioning not only of soil microorganisms but also of soil catalysts, including enzymes.

SUMMARY

The antiseptics toluene and chloroform had little, if any, inhibiting action on the hydrolysis of urea in the soils studied. Carbon disulfide and 50 per cent alcohol as antiseptics likewise allowed considerable hydrolysis to take place.

A hypothesis to explain these and other data envisioned the enzyme urease adsorbed on the soil colloids. In support of such a concept, the natural soil catalytic activity was inactivated or greatly reduced by preheating the moistened soil to 85°C.; by pretreating the soil with the known urease inhibitors: HgCl_2 , hydroquinone, and catechol; and by predigesting the soil with the proteolytic enzyme trypsin, known to destroy the activity of the protein urease in a very short time.

Neither the natural activity nor that of added laboratory Jack-bean urease could be eluted from the soil by any of the substances commonly used for that purpose. The laboratory urease was moderately adsorbed from solution by preheated Yolo fine sandy loam in a few different types of procedure.

Chance observations suggested that the natural soil activity was much more resistant to decomposition than was that of added laboratory urease. Quantitative studies confirmed these observations. If the activity of the soil is due to an enzyme, urease, it must be associated with some other material which gives it some protection against decomposition. It is suggested that a lignoprotein might satisfy such requirements.

REFERENCES

- (1) CONRAD, J. P. 1940 Hydrolysis of urea in soils by thermolabile catalysis. *Soil Sci.* 49: 253-263.
- (2) CONRAD, J. P., AND ADAMS, C. N. 1939 Determining by plant response the retention of nutrient ions by soils. *Jour. Amer. Soc. Agron.* 31: 29-34.
- (3) CONRAD, J. P., AND ADAMS, C. N. 1940 Retention by soils of the nitrogen of urea and some related phenomena. *Jour. Amer. Soc. Agron.* 32: 48-54.
- (4) FALK, K. G. 1924 The Chemistry of Enzyme Actions, ed. 2 Chemical Catalog Co., New York.
- (5) FISHER, R. A. 1932 Statistical Methods for Research Workers, ed. 4. Oliver and Boyd, Edinburgh.
- (6) GRABAR, P., AND RIEGERT, A. 1935 Sur la nature de l'uréase; moyen d'en aborder

⁴ By implication, if not by definition, enzymes arise from the previous activity of living cells. Soil microorganisms must be considered as potential formers of any enzymes that may occur in soils.

- l'étude à l'aide de l'ultra filtration fractionnée. *Compt. Rend. Acad. Sci. [Paris]* 200: 1795-1797.
- (7) HAWK, P. B., AND BERGEIM, O. 1937 Practical Physiological Chemistry, ed. 11. P. Blakiston's Son and Co., Philadelphia.
- (8) JACOBY, M. 1916 Über die Adsorption von Ureasen und ihre Wirksamkeit in ungelöster Form. *Biochem. Ztschr.* 74: 93-96.
- (9) JACOBY, M. 1927 Über die Adsorption der Urease durch Cholesterin. *Biochem. Ztschr.* 190: 157-160.
- (10) OPPENHEIMER, C., AND KUHN, R. 1927 Lehrbuch der Enzyme. G. Thieme, Leipzig.
- (11) PETERS, J. P., AND VAN SLYKE, D. D. 1932 Quantitative Clinical Chemistry Methods. Williams & Wilkins Co., Baltimore.
- (12) QUASTEL, J. H. 1933 The action of polyhydric phenols on urease; the influence of thiol compounds. *Biochem. Jour.* 27: 1116-1122.
- (13) RUSSELL, E. J. 1937 Soil Conditions and Plant Growth, ed. 7. Longmans, Green and Co., London.
- (14) SUBRAHMANYAN, V. 1927 Biochemistry of water logged soils: II. The presence of a deaminase in water logged soils and its role in the production of ammonia. *Jour. Agr. Sci.* 17: 449-467.
- (15) SUMNER, J. B., AND KIRK, J. S. 1932 Über die chemische Natur der Urease. *Ztschr. Physiol. Chem.* 205: 219-230.
- (16) VAN SLYKE, D. D., AND CULLEN, G. E. 1914 The mode of action of urease and of enzymes in general. *Jour. Biol. Chem.* 19: 140-180.
- (17) WAKSMAN, S. A., AND IYER, K. R. N. 1932 Contribution to our knowledge of the chemical nature and origin of humus: I. On the synthesis of the "humus-nucleus." *Soil Sci.* 34: 43-70.
- (18) WAKSMAN, S. A., AND IYER, K. R. N. 1933 Contribution to our knowledge of the chemical nature and origin of humus: IV. Fixation of proteins by lignins and formation of complexes resistant to microbial decomposition. *Soil. Sci.* 36: 69-82.
- (19) WALDSCHMIDT-LEITZ, E., AND STEIGERWALDT, F. 1931 Über die chemische Natur der Urease. *Ztschr. Physiol. Chem.* 195: 260-266.
- (20) WRIGHT, C. H. 1934 Soil Analysis: A Handbook of Physical and Chemical Methods. Thomas Murby & Co., London.

RELATIVE NODULATION OF VARIETIES OF *MEDICAGO SATIVA* VARYING IN SUSCEPTIBILITY TO ALFALFA WILT¹

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In a recent study² of the relative effectiveness of *Rhizobium meliloti* cultures isolated in Kansas, marked variation was noted among the different strains, both in infectivity and in nitrogen-fixing efficiency. In that study Kansas Common alfalfa was employed as the host because it is the variety most widely grown in Kansas. Burton and Wilson³ have noted definite host plant specificity for rhizobia among species of *Medicago* and obtained some evidence of host plant specificity among varieties of *Medicago sativa*. Because of the widespread damage to alfalfa caused by wilt, much effort is being expended in a search for wilt-resistant varieties and strains of *Medicago sativa*. Resistance of the alfalfa plant to infection by *Phytomonas insidiosus* is probably of primary importance in determining its value as a wilt-resistant variety or strain. This suggested the possibility that resistance to infection with *Phytomonas insidiosus* might be associated with resistance to infection with *Rhizobium meliloti* and hence would be reflected in nodulation following inoculation.

METHODS AND MATERIALS

The methods employed were identical with those previously described.² Briefly, they were as follows: The alfalfa seeds⁴ were sterilized, germinated on sterile nutrient agar, and transferred to large cotton-plugged test tubes containing sterile washed Ottawa sand. Prior to sterilization, as much modified Crone's solution, less 15 cc., was added to the sand as it would retain against gravity. The use, as the inoculum for each tube, of 15 cc. of a heavy suspension of young cultures of the rhizobia grown on the Albrecht-McCalla⁵ agar medium, ensured an excess of organisms. The tubes were transferred to the greenhouse and were not opened until the end of the experiment.

¹ Contribution No. 196, department of bacteriology.

² KROULIK, J. T., AND GAINNEY, P. L. Physiological studies of *Rhizobium meliloti* with special reference to the effectiveness of strains isolated in Kansas. *Jour. Agr. Res.* (in press).

³ BURTON, J. C., AND WILSON, P. W. 1939 Host plant specificity among the *Medicago* in association with root-nodule bacteria. *Soil Sci.* 47: 293-301.

⁴ The alfalfa seeds used in these tests were obtained from the department of agronomy, Kansas Agricultural Experiment Station, through C. O. Granfield, assistant agronomist, U. S. Department of Agriculture, in charge of alfalfa wilt investigations in Kansas.

⁵ ALBRECHT, W. A., AND MCCALLA, T. M. 1937 A new culture medium for rhizobia. *Jour. Bact.* 34: 455-457.

When the plants in the better cultures attained a height of 4 to 5 inches, the sand was carefully washed from the roots of all cultures, the number of

TABLE 1

Nodulation and fixation of nitrogen by different strains of Rhizobium meliloti with different varieties of Medicago sativa as host plant—experiment 1

VARIETY OF ALFALFA	STRAIN OF RHIZOBIUM MELILOTI									
	A			23f			208 III			Mean number of nodules per plant, all strains
	Number plants per culture	Number nodules per plant	Nitrogen fixed per plant	Number plants per culture	Number nodules per plant	Nitrogen fixed per plant	Number plants per culture	Number nodules per plant	Nitrogen fixed per plant	
			mgm.			mgm.			mgm.	mgm.
Kansas Common	7	7.7	0.60	8	5.8	0.39	6	18.5	0.39	
	4	8.3	0.71	7	6.1	0.44	6	24.0	0.16	
	7	4.0	0.47	8	5.6	0.41	7	23.9	0.30	
Mean.....		6.7	0.59		5.8	0.41		22.1	0.28	11.5 0.43
Kansas Common (wilt-resistant selection)	8	6.9	0.50	6	6.5	0.55	6	21.0	0.14	
	7	6.1	0.47	8	5.5	0.48	8	25.4	0.34	
	8	5.4	0.50	8	8.3	0.51	8	20.9	0.25	
Mean.....		6.1	0.49		6.8	0.51		22.4	0.24	11.8 0.41
Ladak	8	12.1	0.63	8	8.5	0.35	7	12.0	0.61	
	7	6.7	0.55	8	10.3	0.41	7	13.0	0.44	
	7	7.1	0.81	8	11.0	0.49	7	10.4	0.67	
Mean.....		8.6	0.66		9.9	0.42		11.8	0.57	10.1 0.55
Turkestan	8	6.1	0.63	8	8.6	0.46	8	20.5	0.56	
	7	5.3	0.57	7	9.1	0.40	8	14.0	0.46	
	6	6.0	0.68	8	6.6	0.41	8	17.4	0.48	
Mean.....		5.8	0.63		8.1	0.42		17.3	0.50	10.4 0.52
Turkestan (wilt-resistant selection)	8	4.4	0.48	8	5.3	0.41	7	17.0	0.22	
	8	4.6	0.39	7	5.1	0.44	5	31.4	0.37	
	7	6.7	0.55	7	5.6	0.44	7	24.0	0.33	
Mean.....		5.2	0.47		5.3	0.43		24.1	0.31	11.5 0.40
Mean for all varieties.....		6.5	0.57		7.2	0.44		19.6	0.38	

Differences necessary for significance between means of nitrogen fixed on different varieties, 0.038 mgm.; and between means of different strains, 0.030 mgm.

plants and nodules recorded, and the oven-dried plant material weighed and analyzed for total nitrogen.

TABLE 2

Nodulation and fixation of nitrogen by different strains of Rhizobium meliloti with different varieties of Medicago sativa as host plant—experiment 2

VARIETY OF ALFALFA	STRAIN OF RHIZOBIUM MELILOTI									
	208 III			195 III			A			Mean number of nodules per plant, all strains
	Number plants per culture	Number nodules per plant	Nitrogen fixed per plant	Number plants per culture	Number nodules per plant	Nitrogen fixed per plant	Number plants per culture	Number nodules per plant	Nitrogen fixed per plant	
			mgm.			mgm.			mgm.	mgm.
Kansas Common	8	26.8	0.14	7	39.3	0.09				
	8	30.5	0.22	8	43.1	0.23				
	8	35.8	0.16	8	39.0	0.12				
	8	27.3	0.07	7	25.0	0.08				
Mean		30.0	0.15		36.6	0.13				33.4 0.14
Kansas Common (wilt-resistant selection)	7	30.5	0.08	8	23.9	0.44				
	7	32.6	0.20	8	20.6	0.23				
	7	30.4	0.30	8	26.0	0.04				
	8	25.2	0.27	7	23.9	0.08				
Mean		29.7	0.21		23.6	0.20				26.7 0.21
Ladak	8	31.1	0.49	8	24.8	0.38	8	9.8*	0.60*	
	8	31.9	0.44	8	30.8	0.33	8	7.8	0.69	
	7	25.7	0.53	8	22.9	0.62	8	11.4	0.65	
	8	21.8	0.36	6	24.0	0.30	7	7.4	0.67	
Mean		27.6	0.46		25.6	0.41		9.1	0.65	26.6 0.44
Turkestan	8	27.1	0.49	8	31.9	0.47				
	8	29.1	0.44	7	30.4	0.17				
	8	28.0	0.16	8	27.6	0.30				
	7	19.9	0.33	8	21.9	0.32				
Mean		26.0	0.36		28.0	0.32				27.0 0.34
Turkestan (wilt-resistant selection)	8	47.3	0.29	8	30.4	0.23				
	8	30.9	0.35	8	24.8	0.25				
	8	35.9	0.16	8	25.3	0.22				
	9	25.9	0.23	8	29.7	0.16				
Mean		35.0	0.26		27.6	0.22				31.3 0.24
Mean for all varieties		29.7	0.29		28.3	0.25				

Differences necessary for significance between means of varieties, 0.065 mgm. Differences between means of strains are not significant.

* These cultures were included to enable comparison with an efficient strain and are not included in the analysis of the data.

Three varieties of *Medicago sativa*—Kansas Common, Ladak, and Turkestan—were tested as host plants. Kansas Common is regarded as highly susceptible to wilt; Turkestan, on the other hand, is a highly resistant variety; and Ladak occupies an intermediate position between Kansas Common and Turkestan. In addition to these three varieties, a highly resistant selection of Kansas Common, equally as resistant to wilt as Turkestan, and a selection of Turkestan, representing the most marked resistance to the wilt of any plants available, were also included. In the analysis of the data both these strains are treated as varieties.

Four strains of *Rhizobium meliloti* were tested for infectivity and efficiency in fixing nitrogen on the different varieties of alfalfa. These strains had been previously tested on Kansas Common with the following results: strain A, infectivity low, efficiency high; strain 23f, infectivity low, efficiency average; strain 208 III, infectivity high, efficiency low; strain 195 III, infectivity high, efficiency variable.

Experiment 1 was started October 9 and completed December 11, 1939. The pertinent data collected are recorded in table 1. Experiment 2 was started December 15, 1939, and finished February 23, 1940. The data of interest are recorded in table 2.

DISCUSSION

In the data presented in tables 1 and 2 there is little if any, evidence of an association between varietal susceptibility to alfalfa wilt and infection with *Rhizobium meliloti* as reflected in the number of nodules formed. In experiment 1 the mean numbers of nodules formed on the most susceptible, Kansas Common, and on the most resistant, selected Turkestan, varieties were identical. Also the strains of both Kansas Common and Turkestan selected for resistance to wilt exhibited slightly higher nodulation than did the original varieties. In experiment 2 nodulation is slightly lower in the more wilt-resistant varieties, but again the most wilt-resistant variety, selected Turkestan, ranks next to the most wilt-susceptible variety, Kansas Common, in the number of nodules.

The mean numbers of nodules formed per plant as well as the nitrogen fixed by all strains of rhizobia in symbiosis with the different varieties of alfalfa are indicated in the following summary:

	KANSAS COMMON	KANSAS COMMON (SELECTED)	LADAK	TURKESTAN	TURKESTAN (SELECTED)
Number of nodules.....	21.80	18.80	17.90	18.20	20.80
Nitrogen fixed..... <i>mgm.</i>	0.29	0.32	0.49	0.43	0.32

The data recorded in tables 1 and 2 substantiate previous findings relative to the interrelationship of the four strains of *Rhizobium* and Kansas Common alfalfa, in that strains A and 23f exhibited relatively low infectivity and high

nitrogen-fixing efficiency, whereas strains 208 III and 195 III were highly infective and low in efficiency, the latter showing marked variability. A similar relationship held for the efficient strains A and 23f when other varieties of alfalfa were employed as host plants. The poorer strains, 208 III and 195 III, exhibited a much higher efficiency in fixing nitrogen when Ladak and Turkestan were used as hosts than when Kansas Common was employed. In experiment 1 the number of nodules produced by strain 208 III on Ladak was unusually low for this strain, being only about half as many as were produced upon Kansas Common.

The mean fixation of nitrogen by all strains was significantly higher on Ladak and Turkestan than on either strains of Kansas Common or on se-

TABLE 3

Summary of analysis of variance of data on nitrogen fixation presented in tables 1 and 2

VARIANCE DUE TO	DEGREES OF FREEDOM	SUM OF SQUARES	VARIANCE	F	5 PER CENT LEVEL	1 PER CENT LEVEL	SIGNIFICANCE
<i>Experiment 1</i>							
Variety of <i>Medicago sativa</i>	4	0.2504	.0626	9.725	2.69	4.02	++
Strain of <i>Rh. meliloti</i>	2	0.2949	.1475	22.896	3.32	5.39	++
Variety x strain	8	0.1863	.0233	3.610	2.27	3.17	++
Error	30	0.1933	.0064				
<i>Experiment 2</i>							
Variety of <i>Medicago sativa</i>	4	0.4225	.1056	8.200	3.32	5.39	++
Strain of <i>Rh. meliloti</i>	1	0.0106	.0106	0.822	2.69	4.02	-
Variety x strain	4	0.0018	.0005	0.035	2.27	3.17	-
Error	30	0.3862	.0129				

If *F* exceeds the 5 per cent level, the chances are 9:1 that the observed differences did not arise from experimental error. If *F* exceeds the 1 per cent level, the chances are 99:1 that the variation did not arise from chance, i.e., highly significant (++).

lected Turkestan. This would indicate that the variety of alfalfa may markedly influence the effectiveness of different strains of *Rhizobium*.

The data were submitted to an analysis of variance to determine statistically whether the variations in means of nitrogen fixed were significant. A summary of this analysis is recorded in table 3.

It is evident from this analysis that both the strain of *Rhizobium* and the variety of alfalfa influenced to a highly significant degree the quantity of nitrogen fixed in experiment 1. Furthermore, variance due to the interrelation of strain of organism and variety of plant was highly significant in this experiment.

In the second experiment highly significant differences in nitrogen fixed due to the variety of alfalfa are evident, but differences due to the strain of *Rhizobium* are small. Since only selected low efficiency strains of *Rhizobium*

were included in this experiment, it could hardly be expected that significant differences would be obtained in the fixation of nitrogen. Probably for the same reason, differences in fixation of nitrogen due to interrelation of strain of *Rhizobium* and variety of alfalfa were not significant in experiment 2.

A similar analysis was made of the data from experiment 1 to determine the influence of strain of organism and variety of plant upon the number of nodules formed. Highly significant differences in nodulation due to the strain of organism were evident, a fact established in a previous report, but differences due to variety of alfalfa or to the interrelation of strain of organism and variety of plant were not significant.

SUMMARY

No evidence was obtained indicating an association of resistance in *Medicago sativa* to infection with *Rhizobium meliloti* and resistance to infection with *Phytophthora insidiosa*.

Evidence is submitted substantiating previously reported data from this and other laboratories that highly significant differences in infectivity may be expected to arise from inoculating *Medicago sativa* with different strains of *Rhizobium meliloti*; that highly infective strains are relatively inefficient in fixing nitrogen; and that highly significant differences in the quantity of nitrogen fixed may result when different strains of *Rhizobium meliloti* are employed on the same variety of alfalfa or when the same strain of *Rhizobium meliloti* is employed for inoculating different varieties of alfalfa.

EFFECT OF TREATING DIFFERENT HORIZONS OF SASSAFRAS LOAM ON ROOT DEVELOPMENT OF RED CLOVER¹

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It is obvious that the efficiency with which plant roots occupy the soil has an important bearing on the development of the plant. Horizons which are not penetrated by the root system are of relatively little value from the standpoint of providing moisture or nutrients. In spite of some very notable work on root systems by a small group of investigators, little is yet known of the normal development of roots of various crop plants in soils of the major agricultural regions of the country. This serious gap in information regarding the most effective utilization of soil must be closed if the findings already available on soil conditions and on the growth of the above-ground parts of crops are to be fully utilized.

REVIEW OF LITERATURE

The work of Weaver (47) and his associates and students has given a much better picture of normal root development in the chernozem and prairie soils than is available for any other section of the country. Such information as has been supplied, however, by Goff (24), Booth (4), and Farris (22) indicates that root development in the more humid regions is quite different from that in the natural grassland soils. Whereas the root system of corn was found to extend 6 to 8 feet in depth and an equal distance laterally in eastern Nebraska, Farris observed in New Jersey that the root system rarely extended below 20 inches, and that the lateral spread was not much greater than 3 feet. Similarly, red clover showed a maximum penetration of 9 feet and a lateral spread of about 2½ feet in the upper horizon in eastern Nebraska, which is in sharp contrast with the maximum depth of 18 inches and a lateral spread of less than 1 foot, observed by Farris in New Jersey.

Since observations in the same crop indicated that the soil conditions are directly responsible for the great difference in root development in the humid soils and in the grassland soils, it is of some interest to determine the specific factors involved. No attempt need be made here to cover the entire literature on this field, since it has been satisfactorily treated by Miller (37) for papers prior to 1915. In some cases the investigations were made primarily to determine the general character of the root systems of individual plants.

Investigations of Headden (27) have shown that alfalfa roots had penetrated to depths of 11 to 12 feet in stiff clay soil in a growth period of 6 years, and Cottrell (18) observed penetration of alfalfa roots to a depth of more than 10 feet on 8-year-old plants. Weaver and Crist (48) observed, however, that in the Great Plains where a calcareous hardpan at depths of 1½ to 3 feet occurred, root development of most plants was confined to the layer above the hardpan. Working on a heavy New Jersey soil, Allison (1) found that the response of barley

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to soil aeration was decidedly positive, and concluded that any increase in the imperviousness of the soil might be considered injurious to plant growth through the consequent decrease in the supply of available oxygen. Fitzpatrick and Rose (23) report that depth of root penetration was greatest in nonclaypan soils, intermediate in semiclaypan soils, and least in claypan soils. This difference was attributed to the poor drainage and aeration of the soil immediately above the claypans.

Cannon (11, 13) has shown that root growth is reduced either by lowering the oxygen content of the soil air or by increasing the carbon dioxide content. Root branching was most abundant when air was admitted. Excessive aeration also retarded root development (8). The Howards (29), in India, have reported numerous experiments to substantiate the point that poor soil aeration affects the roots primarily. In heavy soils the roots were confined to the upper soil layers, and in light soils the roots penetrated deeply and widely. Bergman (3) found that plants grown in soil from which oxygen was excluded by flooding showed wilting, etiolation, and loss of leaves. Clements (14) likewise observed that a deficient oxygen supply is detrimental to root development and that this might occur when there is an increase of carbon dioxide in the soil. Bushnell (6) concluded, from growth of potato roots on tiled land, that the increased aeration from the tiles, even when no excess water was present, greatly improved root growth and crop yields.

Kiesselbach (30) concluded that, in general, root development varies inversely as the soil water, and that plants which have their early growth in moderately dry soil may be expected better to withstand a later period of drought because they would have a greater absorbing surface exposed to the soil particles. In the Great Plains, Weaver (47) found that water content of the soil was a controlling factor in root development. Soil texture exerted its effect primarily through water content and aeration. In a more humid climate, however, soil moisture might conceivably be less important than aeration. Shantz (41) has suggested that in humid eastern soils the subsoil² is so fine and colloidal that it takes water very slowly, and may, even when in a seemingly moist condition, contain no water available to plants. In spring and fall, however, moisture is normally present in the subsoil above the wilting point. Depletion of subsoil moisture may be produced by roots of alfalfa in chernozem soils to the extent that roots cannot utilize these horizons until the moisture supply is replenished, according to Kiesselbach et al. (31) and Metzger and Grandfield (34). Hendrickson and Veihmeyer (28) have shown that roots will not grow into soil which contains less moisture than the permanent wilting coefficient.

It is well known that plant roots have definite temperature requirements for optimum growth. Cannon (9) found that root growth may proceed when soil temperatures are favorable, even though atmospheric temperatures are too low for active growth of tops. In general, he (10) noted that roots of plants which characteristically grow deeply or which usually grow in fine-textured soil are less dependent on temperature than are plants with superficial roots. With some species, Cannon (12) found that growth of plants stopped when temperature was below optimum and the soil air was deficient in oxygen, but that growth was resumed at optimum temperatures even when the oxygen supply was unchanged.

Since organic matter is directly concerned with improved soil structure and the accompanying air and moisture relations, it is not surprising to note that Weaver, Hogen, and Weldon (49) report a direct correlation between root growth and the amount of soil organic matter in the various horizons. Washko (45) also found that soybean yields and root development were directly related to soil organic matter.

Watenpugh (46), in studies on alfalfa, observed that root development and yield were correlated definitely with pH and the replaceable calcium of the soil horizons. A pH of 4.8 checked root growth almost entirely, but roots grew well at pH values above 5.0. The rawness of subsoils has been studied by various workers, with contrasting results. Alway, McDole, and Rost (2) ascribe some of the low fertility of eastern Nebraska subsoils to lack

² Throughout the present paper, including the literature review, the term "subsoil" is used, not in its pedological sense, but rather to denote that part of the soil below plow depth.

of nitrogen, and assert that frequently legumes can be grown well on such horizons. Non-legumes were grown successfully on these subsoils when nitrogen was added. On Minnesota glacial soils, however, Harmer (25) found that rawness of subsoil was not always due to lack of nitrogen. McMiller's (33) observations indicated that deficiencies in available phosphates and potassium were responsible for low fertility of those subsoils which were not improved by nitrogen additions. Millar (35) likewise observed that the addition of nitrogen fertilizer did not increase growth of crops on some types of subsoil. Van der Merwe (44) observed that a deficiency of available phosphates was a most important factor limiting the fertility of subsoils and that correction of this deficiency frequently permitted a response to nitrogen additions. Conner (15, 16, 17) found that Indiana subsoils were most likely to be deficient in nitrogen, phosphorus deficiencies being nearly as important. The phosphorus deficiency increased with depth, in spite of the fact that total phosphorus was greater in many of the lower horizons. Exposure of the lower horizons to the action of air greatly increased the availability of the phosphorus present. Millar's (36) work on Hillsdale sandy loam from Michigan showed that the A_2 and B horizons were similarly deficient in available nitrogen and phosphorus, and corn grown on soil from these horizons was greatly stimulated when these deficiencies were corrected. Thornton (43) concluded from an examination of 450 soils in the Middle West that subsoils were generally much lower in available phosphorus but that available potassium was nearly as high in the subsoil as in the surface soil.

Weaver, Jean, and Crist (50) were able to demonstrate that root development of plants is much more profuse in horizons well supplied with nutrients than in those with limited fertility. When roots occupied fertilized layers, their penetration into lower horizons was retarded. They conclude that fertilizer applied solely to the plow zone definitely limits the extension of the plant root system into lower horizons and makes the crop more susceptible to drought because of the reduced volume of soil occupied. Dean (21), in New York, has reported that subsoil application of fertilizer induced deeper rooting of potatoes. Davis (20) observed that the taproot of alfalfa was less extensive on soils fertilized in the A_p horizon than on those not treated. Crist and Weaver (19) asserted that absorption of nutrients from horizons below the A_p materially increases the quantity and quality of crop yields, even when the surface foot is abundantly supplied with a similar nutrient. Haskell (26) also observed that crops responded to applications of plant foods other than potash, when grown on horizons below the A_p . Bushnell (7) reported that potato root development was increased by mixing manure with the subsoil. In New Jersey, Prince, Toth, and Blair (40) tested a large number of intensively farmed soils and found the subsoils to be much lower in total nitrogen, organic carbon, and potassium but higher in total phosphorus than were the topsoils. Tiedjens (42) observed a very beneficial response of asparagus to subsoiling when lime and superphosphate were incorporated with the horizon just below the A_p . In the dry season of 1939, such treatment greatly increased the resistance of the plant to low rainfall, presumably because of the greater soil volume utilized by roots for moisture. Nissley (39), working on similar soils, showed that tillage to 9 inches below plow depth increased the rate of penetration of rainfall into the subsoil and induced deeper penetration of roots and greater tolerance of the crop to limited rainfall.

EXPERIMENTAL METHODS

In an effort to identify more accurately the specific soil characteristics responsible for the limited root development of crop plants in the gray-brown forest soils of New Jersey, an experiment was initiated in September, 1938, in which treatments were applied to Sassafras loam. The plan called for a comparison between a series of treatments made to the plow zone (A_p horizon), and a parallel series in which similar materials were incorporated with the A_2 horizon at a depth of 8-14 inches.

The field was plowed to a depth of 8 inches, smoothed, and two test series

were laid off approximately 150 feet long and 3 feet wide, parallel to each other, about 2 feet apart. The strips were divided into plots 3 by 15 feet, giving 10 plots in each strip. On the strip to be treated in the A_2 horizon the topsoil was removed and placed to one side, and the materials specified in table 1 were applied on top of the horizon and mixed with the soil in the 8-14-inch level with a rotary tilling machine. Following this treatment, the A_p horizon was returned to the area. On the adjoining strip, similar materials were added to the top of the plowed soil and also incorporated with a tilling machine to a depth of about 8 inches.

After these differential treatments had been applied the entire area was uniformly fertilized with 4-12-6 fertilizer, at the rate of 400 pounds per acre, and seeded to wheat on October 13, 1938. Timothy was planted with the wheat in accordance with the common practice of this region. On April 4, 1939, an improved strain of red clover (Kentucky Selection 101) was broadcast at the rate of 10 pounds per acre. The wheat crop was harvested in July, 1939, but no yields were recorded, since the wheat was merely used as a nurse crop for establishing the hay seeding.

In October the root systems of the 6-month-old red clover were examined by the method developed by Weaver (47), in which trenches are dug within a few inches of the plant and the entire root system is exposed by carefully removing the soil with a small pick from around the roots. As each root was revealed in this process, its position was plotted to scale on graph paper; these graphs were later inked-in and photographed. Although the smallest roots and all root hairs are lost by this system, it is possible to observe and plot accurately all the primary and secondary roots of a plant such as red clover. Two typical plants, chosen on the basis of their top growth, were examined in each plot. Since the duplicates were very similar in habit and extent of root system, two observations per plot are believed to give a fairly adequate index of root development.

At the time of root observations in October, soil samples were taken at depths of 3, 10, 16, and 24 inches for determination of volume weight by the Lebedev (32) soil sampler, for mechanical analysis by the hydrometer method of Bouyoucos (5), and for pH values.

Further observations on root development were made on representative plants in April, 1940, 1 year after seeding of the red clover. Since the dry weather of 1939 produced rather thin stands of clover on the plots, it was relatively simple to isolate typical plants and observe the maximum development of root system of red clover under these conditions.

EXPERIMENTAL RESULTS

Determinations made October 1, 1939, 1 year after the initial treatments were applied, showed a pronounced increase in volume weight in the A_2 horizon over the A_p horizon, the 10-inch depth being used as an index of the second horizon (table 1). The A_p horizon appeared to have been reduced in

volume weight on the A₂ treated series, probably because of the removal of the horizon by spading and its return, which produced a more open structure

TABLE 1

Soil conditions on test field, Sassafras loam, October, 1939, 1 year after treatment

PLOT	TREATED HORIZON*	TREATMENT†	VOLUME WEIGHT AT DEPTH OF			CLAY IN SOIL AT DEPTH OF			pH AT DEPTH OF		
			3 inches	10 inches	24 inches	3 inches	10 inches	16 inches	3 inches	10 inches	16 inches
1	A ₂	Tillage only	gm./cc.	gm./cc.	gm./cc.	per cent	per cent	per cent			
11	A _p		1.39	1.40	1.20	18	20	24	6.1	5.3	5.4
			1.43	1.66	1.61	17	17	27	6.0	5.7	5.7
2	A ₂	Tillage, lime	1.31	1.54	1.62	16	18	18	6.0	6.3	5.6
12	A _p		1.35	1.49	1.60	15	18	17	6.0	6.0	5.9
3	A ₂	Tillage, manure	1.29	1.52	1.44	16	26	33	6.1	5.8	5.6
13	A _p		1.33	1.44	1.52	16	18	29	6.1	5.9	5.1
4	A ₂	Tillage only	1.36	1.45	1.25	15	15	23	6.1	5.8	5.9
14	A _p		1.38	1.47	1.58	19	22	40	6.7	5.3	5.8
5	A ₂	Tillage, fertilizer	1.17	1.31	1.38	15	19	40	6.1	5.5	5.5
15	A _p		1.30	1.32	1.17	15	28	26	6.0	5.6	5.7
6	A ₂	Tillage, lime, manure	1.32	1.45	1.42	16	24	30	6.6	5.9	5.8
16	A _p		1.23	1.38	1.36	16	25	29	6.2	5.2	5.5
7	A ₂	Tillage only	1.27	1.34	1.42	17	18	31	6.2	5.5	5.1
17	A _p		1.22	1.33	1.21	16	26	35	5.9	5.2	5.4
8	A ₂	Tillage, lime, fertilizer	1.14	1.31	1.24	18	18	22	5.9	5.9	5.2
18	A _p		1.28	1.41	1.32	17	20	19	6.4	5.1	5.6
9	A ₂	Tillage, lime, manure, fertilizer	1.19	1.30	1.32	17	19	20	6.0	6.0	5.2
19	A _p		1.30	1.34	1.03	17	16	18	6.0	5.3	5.9
10	A ₂	Tillage only	1.21	1.25	1.38	16	18	24	5.8	5.1	5.1
20	A _p		1.33	1.37	1.48	19	21	26	6.1	5.3	5.3

* A_p horizon extends to depth of 8 inches, A₂ horizon approximately 8-14 inches, and B horizon 14-30 inches.

† Rates of application, per acre: limestone—1 ton, fertilizer—400 pounds of 4-12-6, manure—10 tons.

than was obtained where the soil was handled entirely by ordinary farm practices. In the B horizon, as represented by the samples from the 24-inch layer, the volume weight varied considerably, in some cases being lower than

the A_2 and in some cases higher. This may be attributed to the irregular occurrence of sandy strata in the B horizon.

Differences in clay content of the A_p horizon were relatively small on all the plots. At the 10-inch depth the clay content was substantially greater than in the plow zone, on all except one plot. The clay content of the B horizon was in general higher than that of the A_2 , as would be expected.

On the basis of volume weight and clay content, as index of structural conditions of the soil, it appears that the A_p horizon of the plots on which treatments were applied to the A_2 layer was somewhat more open and easily penetrated than the plow depth of plots receiving no subsoil treatment. The A_2 horizon of the A_2 treated plots had a reduced volume weight in 6 of 10 plots when direct comparisons are made with plots not so treated. These differences were not strongly pronounced, indicating that tillage of the A_2 horizon one year earlier had made no striking changes in porosity. In spite of this fact, the figures on root development in the A_2 treated horizon (table 2) show that substantial improvement in the suitability of this horizon for occupation by roots had occurred.

The surface soil was, in general, only slightly acid, with pH values of approximately 6.0. In the A_2 horizon the pH was somewhat lower, ranging from 5.1 to 5.8. Where lime had been applied in the A_p horizon the pH of that horizon had been improved, but in no case ran higher than 6.4; and lime in the A_2 horizon had substantially raised the pH. There was little or no effect on the 16-inch depth from lime applied 1 year prior to the soil sampling, either in the surface horizon or in the A_2 horizon. Other chemical tests made on the soil samples at various depths indicated surprisingly little difference in available phosphate and potash, as measured by the Morgan (38) method. Since root response to the treatments was obvious fully $1\frac{1}{2}$ years after the initial treatment, it seems likely that the benefits from these treatments were due to factors not detected by these chemical tests.

Effect of treatments on red clover roots 6 months after seeding

One of the most significant responses to soil treatment was that produced by tillage of the A_2 horizon. It may be noted from table 2 that such A_2 tillage produced a substantial increase in both primary and secondary roots in the plow zone itself. The greatest effect, however, was observed in the A_2 horizon receiving tillage only, in which the increased root growth was nearly twice as great as where such tillage had not been given the second horizon.

The effect of tilling the A_2 horizon was found in the 14-30-inch depth as well as in the horizon actually tilled. Thus on plot 4, where the A_2 horizon had been tilled, the total root extent in the B horizon was 102 inches, in contrast with only 20 inches in the same horizon of the paired plot 14, where the A_2 had not been tilled. When the total length of root systems per plant is considered, plots in which the A_2 horizon had been tilled, without any other treatment, produced approximately twice as many roots as the A_p tillage

only, which served as a check. Since a great proportion of this increase was found in the A₂ and B horizons, it is clear that on this particular soil where the A₂ is obviously somewhat more compact and has a higher clay content than the A_p, tillage alone has been an important means of increasing root

TABLE 2
Extent of red clover roots, as affected by soil treatments, in October, 1939, 6 months after seeding
Root length in inches

PLOT	TREATED HORIZON	TREATMENT	TOTAL LENGTH OF CLOVER ROOTS PER PLANT									
			Soil horizon 0-8 inches			Soil horizon 8-14 inches			Soil horizon 14-30 inches			Total all horizons
			Primary	Secondary	Total	Primary	Secondary	Total	Primary	Secondary	Total	
1	A ₂	Tillage only	22	64	86	20	59	79	53	83	136	301
11	A _p		15	35	50	7	12	19	3	5	8	77
2	A ₂	Tillage, lime	28	63	91	13	36	49	7	15	22	162
12	A _p		20	35	55	18	21	39	9	15	24	118
3	A ₂	Tillage, manure	19	56	75	23	36	59	7	12	19	153
13	A _p		15	47	62	15	22	37	2	3	5	104
4	A ₂	Tillage only	16	96	112	17	50	67	38	64	102	281
14	A _p		18	65	83	16	18	34	8	12	20	137
5	A ₂	Tillage, fertilizer	9	40	49	7	19	26	9	27	36	111
15	A _p		13	57	70	5	16	21	3	9	12	103
6	A ₂	Tillage, lime, manure	19	70	89	13	35	48	14	26	40	177
16	A _p		20	45	65	18	26	44	7	26	33	142
7	A ₂	Tillage only	20	88	108	21	55	76	7	33	40	224
17	A _p		18	84	102	5	30	35	1	9	10	147
8	A ₂	Tillage, lime, fertilizer	18	75	93	10	40	50	9	26	35	178
18	A _p		15	66	81	6	15	21	1	7	8	110
9	A ₂	Tillage, lime, fertilizer, manure	16	50	66	15	54	69	13	21	34	169
19	A _p		11	110	121	7	15	22	0	2	2	145
10	A ₂	Tillage only	32	54	86	35	45	80	57	54	111	277
20	A _p		19	65	84	17	32	49	9	19	28	161

occupation of the lower horizon. No measurements were made of any differences in aeration or oxygen-supplying power of these soil horizons, but since the occupation of the B horizon had been greatly improved, even though this stratum was untouched and was characteristically higher in clay content, it

seems likely that improved aeration was the cause of the greater development of the root system, rather than ease of physical penetration of the soil or such chemical characteristics as pH or supply of available nutrients.

When the tillage of the A_2 horizon was accompanied by the addition of lime, manure, or fertilizer, alone or in combination, the response was considerably different. Comparisons made between A_2 treated plots and the check plots 11, 14, 17, and 20 indicate that the addition of any of these materials to the A_2 horizon by tillage had substantially increased root development. Such

TABLE 3

*Comparative effect of tillage versus treatment with lime, manure, or fertilizers, on root development of red clover, 6 months after seeding**

Root length in inches

HORIZON	ROOT LENGTH PER PLANT ON TILLAGE-ONLY PLOTS TREATED HORIZON		ROOT LENGTH PER PLANT ON "FERTILIZED" PLOTS† TREATED HORIZON	
	A_2	A_p	A_2	A_p
<i>0-8 Inches</i>				
Primary roots.....	22	17	18	16
Secondary roots.....	75	62	59	60
Total.....	97	79	77	76
<i>8-14 Inches</i>				
Primary roots.....	23	11	13	11
Secondary roots.....	52	23	37	19
Total.....	75	34	50	30
<i>14-30 Inches</i>				
Primary roots.....	38	5	10	4
Secondary roots.....	58	11	21	10
Total.....	96	16	31	14
Total per plant, all horizons.....	268	129	158	120

* Root observations made in October, 1939, on seeding made April 4, 1939.

† "Fertilized" plots are those treated with lime, manure, or fertilizer, alone or in combination.

development was not so great as when tillage was used alone. Furthermore, when additional lime, manure, or fertilizer had been incorporated in the plow zone, there was even further tendency for the root system to be confined to the plow depth than when such materials were not incorporated or when tillage only was given to the A_2 horizon. Comparison of the relative effectiveness of lime, manure, and fertilizer incorporated in the A_2 horizon indicated that manure and lime were more important than fertilizer, and the combination of all three substances produced the greatest response in root develop-

ment of the treated A_2 . A further important observation is that these materials when present in the A_2 exerted a definite inhibiting effect on occupation of the B horizon, when comparison was made with A_2 tillage plots. There is a clear trend, shown in these root observations made at 6 months, toward an increase in root abundance in the layer which was improved by the incorporation of lime, manure, or fertilizer. There was an extensive development of roots in the plow zone when materials were incorporated in that horizon, and a secondary concentration of roots in the A_2 horizon when materials were incorporated at that depth, in contrast with a tendency for much greater root penetration and occupation of all horizons when tillage only was given the A_2 . It appears that the chemotropic response of red clover roots under these conditions masked to a large extent the response to the assumed increase in aeration which resulted from tilling only.

Since the general effect of lime, manure, and fertilizer was somewhat the same, it seems worthwhile to summarize the effect of tillage versus normal treatment, as given in table 3. Tillage in the A_2 horizon increased the total root length to 268 inches, in contrast with 124 inches where the normal treatment was given only to the A_p . Although the addition of soil-improving materials to the A_2 horizon produced a root length of 158 inches, better distributed through the three horizons than on the check A_p plots, a pronounced trend was evident for such treatments to reduce root development below that of A_2 tillage alone. When the additional treatments were applied only to the surface soil, there was surprisingly little difference from the check plots. This might be taken to mean that the normal soil, which was in reasonably good condition as to acidity and plant food content, was adequate for nearly optimum root development under these conditions.

Effect of treatments on red clover roots 1 year after seeding

The observations made six months later on the root systems of the clover indicate that there had been a very active growth between early October and early April. Since the temperatures were abnormally low during this period (table 6), the increased growth indicated in this experiment is probably less than would be found normally. After allowance is made for discrepancies which are bound to exist when different plants are observed at different periods, it seems obvious that the increase in root length was due principally to development of secondary roots rather than to further extension of primary roots.

In general, the trends noted at 6 months of age were emphasized still further in the 1-year-old clover plants (table 4). Plants in plots which received tillage only in the A_2 horizon continued to have very much better root development than plants in plots which had not been so tilled. This is particularly true when the roots in the A_2 horizon and those in the B horizon were considered. Plants grown on the plots receiving lime, manure, or fertilizer, alone or in combination, in the A_2 horizon, produced substantially more roots

in that horizon than plants not so treated, but the occupation of the B horizon by these plants was relatively limited. There was a strong tendency for root systems to be very well developed in the particular layer receiving lime,

TABLE 4

Extent of red clover roots, as affected by soil treatments, in April, 1940, 1 year after seeding
Root length in inches

PLOT	TREATED HORIZON	TREATMENT	TOTAL LENGTH OF CLOVER ROOTS PER PLANT									
			Soil horizon 0-8 inches			Soil horizon 8-14 inches			Soil horizon 14-30 inches			Total all horizons
			Primary	Secondary	Total	Primary	Secondary	Total	Primary	Secondary	Total	
1	A ₂	Tillage only	29	177	206	25	53	78	20	85	105	389
11	A _p		22	130	152	7	27	34	0	3	3	189
2	A ₂	Tillage, lime	17	169	186	9	63	72	4	12	16	274
12	A _p		18	142	160	10	15	25	2	14	16	201
3	A ₂	Tillage, manure	33	160	193	20	121	141	2	14	16	350
13	A _p		19	210	229	12	21	33	0	22	22	284
4	A ₂	Tillage only	20	191	211	31	119	150	19	76	95	456
14	A _p		22	142	164	6	15	21	0	4	4	189
5	A ₂	Tillage, fertilizer	27	154	181	13	106	119	0	15	15	315
15	A _p		30	150	180	3	17	20	5	6	11	211
6	A ₂	Tillage, lime, manure	20	130	150	8	102	110	0	10	10	270
16	A _p		14	157	171	10	17	27	0	22	22	220
7	A ₂	Tillage only	19	129	148	25	149	174	9	95	104	426
17	A _p		15	127	141	8	10	18	2	11	13	172
8	A ₂	Tillage, lime, fertilizer	24	123	147	17	85	102	1	13	14	263
18	A _p		21	361	382	5	37	42	0	2	2	426
9	A ₂	Tillage, lime, fertilizer, manure	23	124	147	8	131	139	0	5	5	291
19	A _p		21	377	398	7	38	45	0	7	7	450
10	A ₂	Tillage only	18	134	152	15	77	92	6	56	62	306
20	A _p		15	242	257	15	7	22	0	6	6	285

manure, or fertilizer, and failure for plants under such treatment to develop deeper root systems than those under tillage only (figs. 1-8).

Since the B horizon is presumed to be better supplied with total plant nutrients than the A₂ horizon, it is of interest to examine the root development in the B horizon specifically. From this standpoint it is very clear that tillage

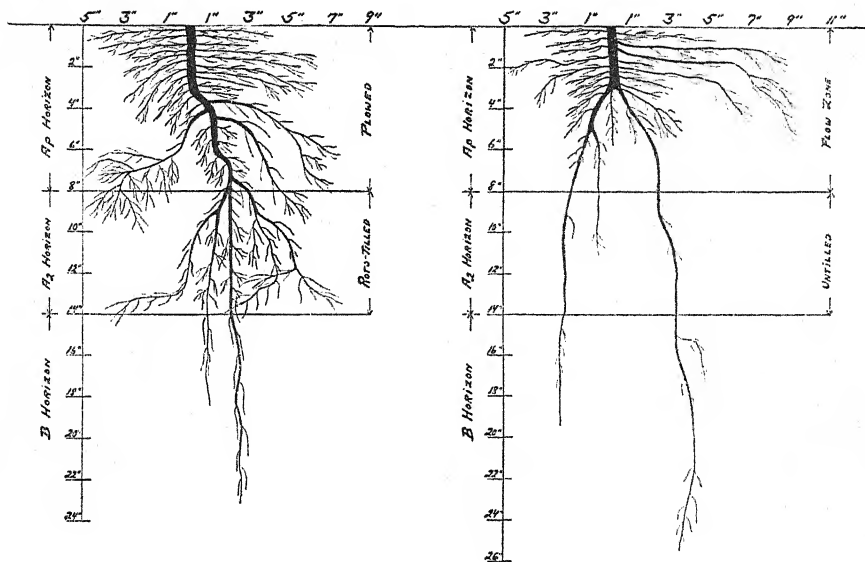


FIG. 1. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 2, plant A. Soil treatment: plow zone, normal fertilizer; subsoil, tillage plus lime. *Right.* Plot 12, plant A. Soil treatment: plow zone, normal fertilizer plus lime; subsoil, untreated.

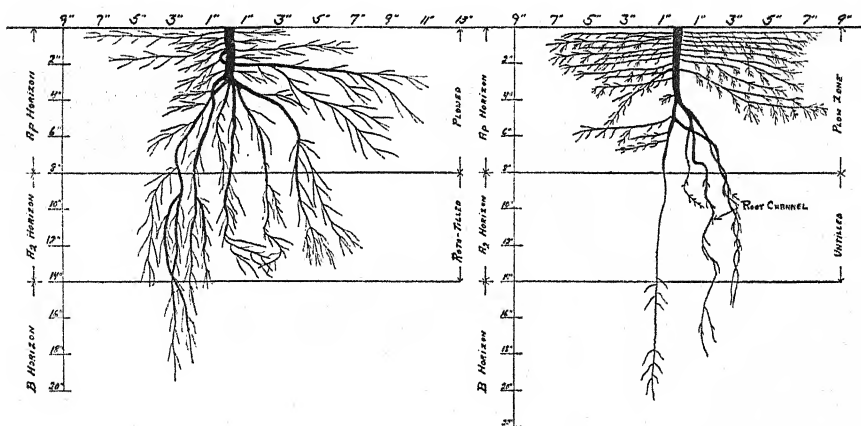


FIG. 2. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 3, plant A. Soil treatment: plow zone, normal fertilizer; subsoil, tillage plus manure. *Right.* Plot 13, plant A. Soil treatment: plow zone, normal fertilizer plus manure; subsoil, untreated.

in the A_2 horizon was the most effective means of inducing root development in the B horizon. Tillage of A_2 , accompanied by lime, manure, and fertilizer, alone or in combination, produced a slight increased occupation of the B

horizon in comparison with A_p plots, but much less than tillage alone. This condition seems somewhat contrary to general observations made by some

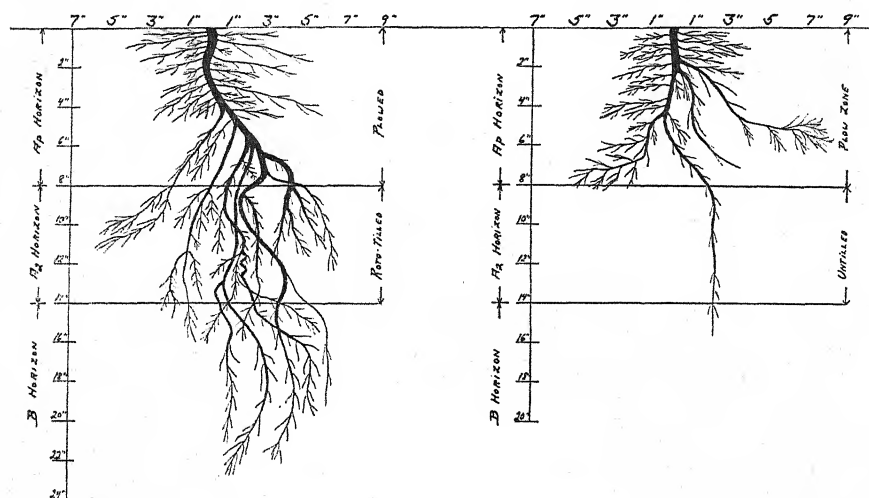


FIG. 3. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 4, plant A. Soil treatment: plow zone, normal fertilizer; subsoil, tillage only.

Right. Plot 14, plant A. Soil treatment: plow zone, normal fertilizer; subsoil, untreated.

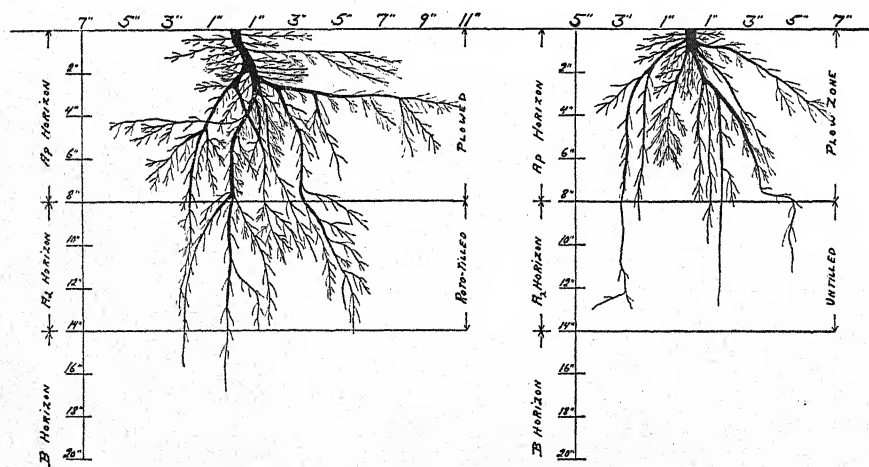


FIG. 4. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 5, plant A. Soil treatment: plow zone, normal fertilizer; subsoil, tillage plus 4-12-6 fertilizer. *Right.* Plot 15, plant A. Soil treatment: plow zone, normal fertilizer plus 4-12-6 fertilizer; subsoil, untreated.

workers that subsoiling and other tillage of the zone below the A_p horizon have comparatively little value unless accompanied by additional soil-im-

proving materials. It may be that the A_2 horizon in this particular test area was considerably different from that of other soils where observations of other workers have been made. Visual examination, however, failed to indicate

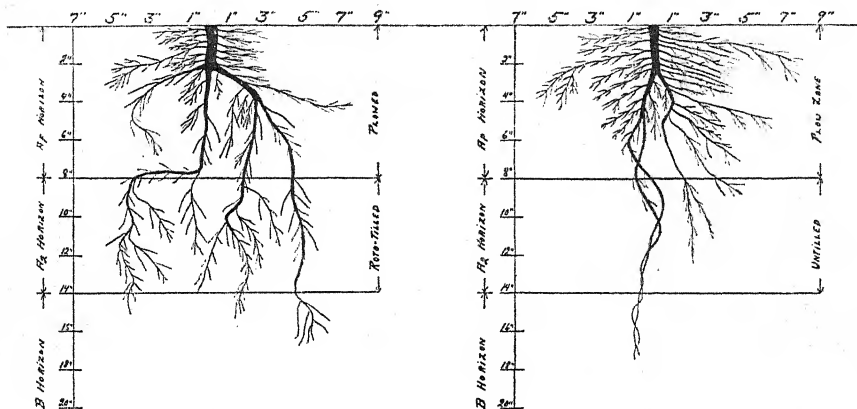


FIG. 5. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 6, plant A. Soil treatment: plow zone, normal fertilizer; subsoil, tillage plus lime and manure. *Right.* Plot 16, plant A. Soil treatment: plow zone, normal fertilizer plus lime and manure; subsoil, untreated.

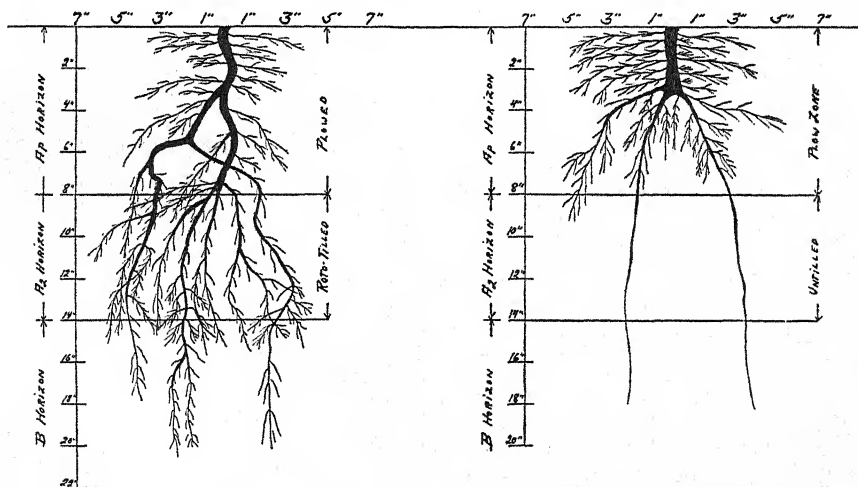


FIG. 6. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 7, plant B. Soil treatment: plow zone, normal fertilizer; subsoil, tillage only. *Right.* Plot 17, plant B. Soil treatment: plow zone, normal fertilizer; subsoil, untreated.

any hardpan or plow sole in this experiment, which was apparently conducted under normal conditions. Since these observations were confined to red clover only, no knowledge is at hand regarding the comparative response of other crop plants to these conditions.

Averages of the results from the four tillage-only plots and of those from the remaining six plots receiving lime, manure, or fertilizer, alone or in combination, show that the A_2 horizon was much more adequately occupied when tillage was given alone than when tillage was given with soil-improving ma-

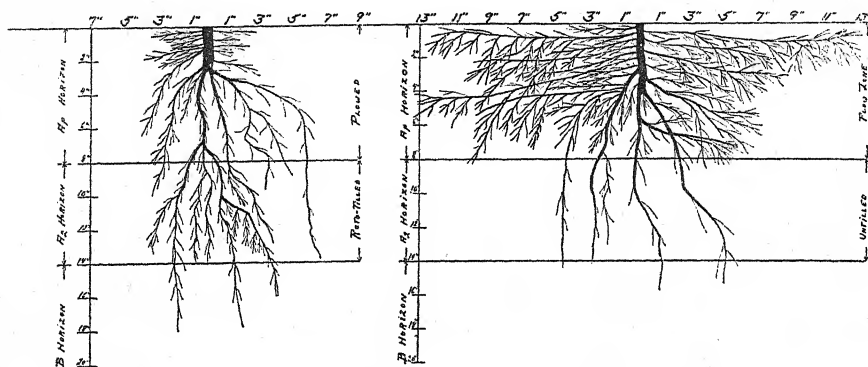


FIG. 7. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 8, plant B. Soil treatment: plow zone, normal fertilizer; subsoil, tillage plus lime and fertilizer. *Right.* Plot 18, plant B. Soil treatment: plow zone, normal fertilizer plus lime and fertilizer; subsoil, untreated.

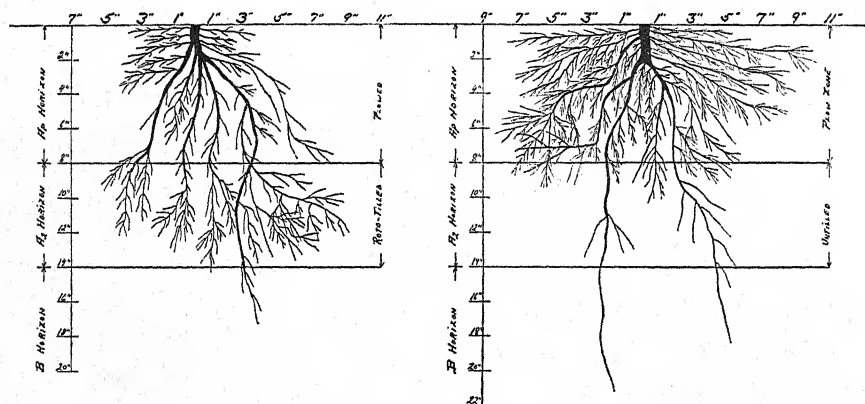


FIG. 8. ROOTS OF RED CLOVER, 1 YEAR AFTER SEEDING

Left. Plot 9, plant A. Soil treatment: plow zone, normal fertilizer; subsoil, tillage plus lime, manure, and fertilizer. *Right.* Plot 19, plant A. Soil treatment: plow zone, normal fertilizer plus lime, manure, and fertilizer; subsoil, untreated.

terials. The tendency for plants to develop root systems rather extensively in zones receiving fertilizer or other plant food is strongly indicated in table 5. The poorest total root development was obtained when the surface soil only was tilled and no treatment was given to any other horizon. It is clear, therefore, that a substantial increase in root development does follow surface soil

improvement, even on Sassafras loam of average to good fertility, and that substantial increase in utilization of the subsoil actually treated occurs when the materials are incorporated below the surface. Since treatments made at any depth seem to have an inhibiting effect on root development below the zone treated, it is possible that the quantities of material used here were sufficient to inhibit more extensive root development through stimulation of development in the treated zone. Possibly lighter applications, accompanied by tillage, would have produced the desired effect of increasing not only the

TABLE 5

*Comparative effect of tillage versus treatment with lime, manure, or fertilizers, on root development of red clover, 1 year after seeding**

HORIZON	ROOT LENGTH PER PLANT ON TILLAGE-ONLY PLOTS		ROOT LENGTH PER PLANT ON "FERTILIZED" PLOTS	
	TREATED HORIZON		TREATED HORIZON	
	A ₂	A _p	A ₂	A _p
<i>0-8 Inches</i>				
Primary roots.....	21	18	24	20
Secondary roots.....	158	160	140	233
Total.....	179	178	164	253
<i>8-14 Inches</i>				
Primary roots.....	24	9	12	8
Secondary roots.....	99	15	101	24
Total.....	123	24	113	32
<i>14-30 Inches</i>				
Primary roots.....	13	1	1	1
Secondary roots.....	78	6	11	10
Total.....	91	7	12	11
Total per plant, all horizons.....	393	209	289	296

* Root observations made in April, 1940.

occupation of the treated zone, but also of root stimulation and further penetration. This point was not tested in this experiment.

Relation of root development to yield of tops

It is of interest to note whether the increased root development produced by tillage of the A₂ horizon or by the incorporation of soil amendments in the A_p or in the A₂ horizon influenced yields of tops. In analyzing such relationships, the possible influence of the weather conditions must be considered. Table 6 shows that the monthly rainfall during 1939 was considerably below normal. This produced a thin stand of clover plants. Beginning in March,

1940, however, rainfall was abundant, and even the plants growing with the least extensive root systems should not have been short of moisture. This is particularly true since the spring of 1940 was cool, preventing any rapid dissipation of moisture. As it is well known that very strong plants can be grown in solution culture or in sand culture with comparatively limited root systems, provided the necessary moisture and nutrients are available, it is not

TABLE 6

Weather conditions during the period covered by this experiment, New Brunswick, New Jersey

MONTH AND YEAR	TEMPERATURE		PRECIPITATION	
	Mean temperature	Departure from normal	Monthly total	Departure from normal
	°F.	°F.	inches	inches
<i>1938</i>				
September.....	63.8	-2.1	10.28	+6.72
October.....	56.2	+1.5	2.30	-1.11
November.....	45.6	+2.0	3.64	+0.46
December.....	34.6	+0.9	2.68	-0.97
<i>1939</i>				
January.....	30.2	-0.3	3.53	-0.04
February.....	37.0	+6.5	5.52	+1.91
March.....	39.7	+0.6	4.42	+0.64
April.....	48.0	-1.7	3.89	+0.28
May.....	63.0	+2.6	0.80	-2.93
June.....	71.2	+2.4	3.48	-0.23
July.....	73.4	-0.3	2.77	-2.02
August.....	75.6	+3.8	4.27	-0.48
September.....	66.7	+0.8	1.81	-1.75
October.....	55.4	+0.7	4.09	+0.68
November.....	42.1	-1.5	1.74	-1.44
December.....	35.5	+1.8	1.13	-2.52
<i>1940</i>				
January.....	22.2	-8.7	1.62	-2.03
February.....	31.9	+1.3	3.11	-0.44
March.....	33.6	-5.6	4.20	+0.43
April.....	45.0	-4.6	5.50	+1.85
May.....	58.5	-1.9	6.74	+3.01

surprising that rapid growth of the first cutting of red clover in 1940 was not strikingly affected by the various treatments.

Results of the first harvest of tops made in June, 1940, are given in table 7. The combination of lime, fertilizer, and manure produced the greatest yields, lime alone being the least valuable of the soil amendments. Tillage of the A₂ horizon produced no substantial increase in growth over that on untreated plots tilled only in the A_p. It seems proper to assume that, even though tillage of the A₂ horizon or the incorporation of soil-improving materials at

that depth did not increase yields in proportion to increased root occupation, a direct correlation might be expected under very unfavorable weather con-

TABLE 7

Yield of first cutting red clover, harvested June 15, 1940, as affected by treating different soil horizons in October, 1938

TREATMENT APPLIED	DRY WEIGHT OF 10 REPRESENTATIVE PLANTS	
	Grown on plots treated in A ₂ horizon	Grown on plots treated in A _p horizon
	gm.	gm.
Tillage only.....	155	200
Tillage, lime.....	227	184
Tillage, manure.....	256	215
Tillage only.....	217	241
Tillage, fertilizer.....	231	279
Tillage, lime, manure.....	288	277
Tillage only.....	278	213
Tillage, lime, fertilizer.....	274	312
Tillage, lime, fertilizer, manure.....	307	386
Tillage only.....	200	181
<i>Averages</i>		
For 4 tillage-only plots.....	212.5	208.8
For 6 treated plots.....	263.8	275.5

ditions such as limited moisture and excessive heat. Further studies on this point are needed.

DISCUSSION

It is evident from this study that the root systems of red clover plants grown on Sassafras loam in New Jersey differ decidedly from those reported by workers in natural grassland regions. Not only do they fail to penetrate the soil to a depth of 4 to 10 feet, as others have reported, but their lateral development is limited. The deepest root observed extended to a depth of 30 inches on a plot which was tilled at the depth of the A₂ horizon.

It is assumed that the soil factors are responsible for the lack of deep penetration by the roots, since the temperature and rainfall in New Jersey are believed to be almost ideal for the growing of clover.

The most important result of this study was the discovery of the good effect of tillage in the A₂ horizon on root penetration and on root occupation of the B horizon immediately below the tilled zone. In the observations on 6-month-old plants, this treatment resulted in an increase of more than 200 per cent in root development in the B horizon over that noted on the next best plots, and an increase of about 500 per cent over the results obtained from the other two types of treatment. In the observations on 1-year-old plants, the B horizon roots from the A₂ tillage plots showed a 700 per cent increase in

growth over those from the A_2 fertilized and A_p fertilized plots and a 1300 per cent increase over those from the A_p tillage plots. None of the physical and chemical tests made at the time of root observations in October, 1939, throw any light on what might have caused this increased root development in the B horizon. Increased aeration, however, seems to offer the most logical explanation. When the A_2 horizon was tilled, it may be assumed that the ventilation of the soil was improved. The difference might be due not only to increased aeration of the entire A_2 horizon but to the breaking up of a plow sole, if one existed. In the process of root examinations, however, the A_2 horizon seemed to have a very open structure and gave no indication of excessive compactness.

Another possible explanation lies in the greater supply of available phosphorus. Tiedjens (42) has reported that the subsoils of New Jersey are depleted of available phosphorus and respond to superphosphate and lime applications. Possibly the phosphorus of the subsoils in the present experiment became more available for plant use as a result of the improved aeration. Conner (17) has shown that although the phosphorus content of the subsoil may be as high as that of the surface soil, its availability is much lower; after the subsoil had been exposed to the air for some time, the availability of the phosphorus was increased.

One point clearly shown is the effect of combinations of lime, manure, and fertilizer in the A_p horizon on root development in that horizon. The response was very marked, roots extending outward as far as 13 inches on each side of the plants. Most of this root development was confined to that horizon, very few of the roots extending even into the A_2 horizon. Where these materials (lime, manure, and fertilizer) were added individually, the root growth was less than where they were used in combination; nevertheless, the root development in the A_p horizon of the former showed a large increase over that in the check plots (A_p tillage) and over that in the A_2 fertilized plots. The A_2 fertilized plots showed a marked increase in root development in the A_2 horizon over the check plots (A_p tillage) and the A_p fertilized. The addition of fertilizer materials to the A_p horizon, however, seemed to retard root development into the B horizon.

The results from all plots that had received soil amendments, A_p or A_2 , showed root stimulation similar to that observed by Weaver (47) and others. These workers found that additions of fertilizer tended to concentrate root development in the region where the soil amendment was placed, development outside that region being much less. This is clearly shown in the A_2 fertilized plots. Besides soil amendments, these plots also received tillage similar to that of the A_2 tillage plots. The results of this study show that with tillage alone in the A_2 horizon the roots penetrate far into the B horizon, but the addition of fertilizer causes the roots to concentrate in the fertilized zone.

The A_p tillage plots, which were used as check plots, show the normal development of red clover roots in New Jersey. These results correspond with the findings of Farris (22). In these plots root development was most ex-

tensive in the A_p horizon, decreased sharply in the A_2 , and almost ceased in the B.

In the A_p fertilized plots, the combined use of lime and fertilizer and of lime, manure, and fertilizer resulted in increases in total root development over those with lime, manure, or fertilizer used alone. In the A_2 fertilized plots, however, this was not found to be true, the response to all soil amendments being of similar order.

Another important fact shown by this study was the marked increase in total root development of the plants of the A_2 tillage plots over that in all other plots. Six months after seeding, the A_p tillage plots had an average total length of roots of 129 inches; the A_p fertilized plots, 120 inches; the A_2 fertilized plots, 158 inches; and the A_2 tillage plots, 268 inches. One year after seeding, the A_p tillage plots had an average total root length of 209 inches; the A_2 fertilized plots, 289 inches; the A_p fertilized plots, 296 inches; and the A_2 tillage plots, 393 inches.

Whereas Washko (45), in studying soybeans under New Jersey conditions, found that, when the soil was not greatly lacking in any of the ordinary nutrient elements, a lack of soil organic matter was the limiting factor in root development, Farris (22) concluded that this factor influences root systems to only a limited extent, since the roots in his experiments with red clover penetrated into soil horizons which contained very little organic matter. These observations favor the conclusions drawn by Farris.

CONCLUSIONS

This study shows that root penetration and occupation of the B horizon increased when the A_2 horizon received tillage only. It also shows that plants from A_2 tillage plots had a greater total root development than those from any of the other treatments. When fertilizer, lime, and manure were placed in the A_p horizon, the total root development increased but was restricted to the A_p horizon. From this, one might conclude that when commercial fertilizers are concentrated in the plowed depth of soil, plant roots will tend to restrict their growth chiefly to that horizon.

SUMMARY

The root system of red clover was studied after growing in Sassafras loam with various cultural and fertilizer treatments, in order to determine the limiting factor or factors of root development in New Jersey.

The root systems of all red clover plants examined were less extensive than those reported in the western and midwestern states.

The chemical and physical properties of the soil studied; namely, soil acidity, texture, pore space, and nutrient supply, apparently had little influence on root penetration.

The plots in which occurred the deepest penetration and occupation of the B horizon by roots were those tilled in the A_2 horizon without other treatment.

Those plots which received combinations of lime, manure, and fertilizer

had an extensive root development in the A_p horizon, a much more limited one in the A_2 horizon, and almost none in the B horizon.

This study indicates that some factor or factors in the A_2 horizon of certain New Jersey soils is corrected by tilling, and that this permits roots of red clover to pass through this horizon and develop extensively in the B horizon. Additional investigation is needed to find the underlying causes for this response to tillage.

REFERENCES

- (1) ALLISON, R. V. 1924 Effect of aeration upon the development of barley in a heavy clay soil. *Soil Sci.* 17: 97-106.
- (2) ALWAY, F. J., MCDOLE, G. R., AND ROST, C. O. 1917 The loess soils of the Nebraska portion of the transition region: VI. The relative rawness of the subsoils. *Soil Sci.* 3: 9-36.
- (3) BERGMAN, H. F. 1920 Relation of aeration to the growth and activity of roots and its influence on the ecesis of plants in swamps. *Amer. Jour. Bot.* 34: 13-33.
- (4) BOOTH, N. O. 1908 Root development. *N. Y. Agr. Exp. Sta. Ann. Rpt.* 26 (pt. 3): 345-349.
- (5) BOUYOUCOS, G. J. 1930 A comparison of the hydrometer method and the pipette method for making mechanical analysis of soils, with new directions. *Jour. Amer. Soc. Agron.* 22: 747-751.
- (6) BUSHNELL, J. 1935 Sensitivity of the potato plant to soil aeration. *Jour. Amer. Soc. Agron.* 27: 251-253.
- (7) BUSHNELL, J. 1937 Fertilizers in the subsoil for potatoes. *Amer. Potato Jour.* 14: 78-80.
- (8) CANNON, W. A. 1912 Physical relations of roots to soil factors. *Carnegie Inst. Wash. Yearbook* 11: 61-62.
- (9) CANNON, W. A. 1917 Soil temperature and plant growth. *Plant World* 20: 361-363.
- (10) CANNON, W. A. 1918 Modification of root habits by experimental means. *Carnegie Inst. Wash. Yearbook* 17: 83-85.
- (11) CANNON, W. A. 1920 Effect of a diminished oxygen supply in the soil on the rate of growth of roots. *Carnegie Inst. Wash. Yearbook* 19: 59-61.
- (12) CANNON, W. A. 1921 Root growth in relation to a deficiency of oxygen or an excess of carbon dioxide in the soil. *Carnegie Inst. Wash. Yearbook* 20: 48-51.
- (13) CANNON, W. A. 1925 Physiological features of roots. *Carnegie Inst. Wash. Pub.* 368.
- (14) CLEMENTS, F. E. 1921 The role of oxygen in root activity. *Carnegie Inst. Wash. Pub.* 315.
- (15) CONNER, S. D. 1930 Fertility tests of soil horizons. *Amer. Soil Survey Assoc. Bul.* 11: 122-125.
- (16) CONNER, S. D. 1935 Nitrogen, phosphorus and potassium requirements of Indiana surface soils and subsoils. *Jour. Amer. Soc. Agron.* 27: 52-56.
- (17) CONNER, S. D. 1936 Relative fertility of surface soils and their subsoils. *Purdue Agr. Exp. Sta. Ann. Rpt.* 49: 18.
- (18) COTTRELL, H. M. 1902 Growing alfalfa in Kansas. *Kans. Agr. Exp. Sta. Bul.* 114: 61-62.
- (19) CRIST, J. W., AND WEAVER, J. E. 1924 Absorption of nutrients from subsoil in relation to crop yield. *Bot. Gaz.* 77: 121-148.
- (20) DAVIS, B. M. 1924 Root development of lucerne. *New Zeal. Jour. Agr.* 28: 179-180.
- (21) DEAN, D. 1923 Sub-surface application of potato fertilizers. *Rpt. Potato Assoc. Amer. Meeting* 10: 110-113.
- (22) FARRIS, N. F. 1934 Root habits of certain crop plants as observed in the humid soils of New Jersey. *Soil Sci.* 38: 87-111.

- (23) FITZPATRICK, E. G., AND ROSE, L. E. 1936 A study of root distribution in prairie clay-pan and associated friable soils. *Amer. Soil Survey Assoc. Bul.* 17: 136-145.
- (24) GOFF, E. S. 1888 Influence of depth of tillage upon the depth of roots. *N. Y. Agr. Exp. Sta. Ann. Rpt.* 7: 171-173.
- (25) HARMER, P. M. 1918 Relative rawness of some humid subsoils. *Soil Sci.* 5: 393.
- (26) HASKELL, S. B. 1925 Availability of subsoil potash. *Soil Sci.* 19: 105-114.
- (27) HEADDEN, W. P. 1896 Alfalfa. *Colo. Agr. Exp. Sta. Bul.* 35: 47-59.
- (28) HENDRICKSON, A. H., AND VEIHMAYER, F. J. 1931 Influence of dry soil on root extension. *Plant Physiol.* 6: 567-576.
- (29) HOWARD, A., AND HOWARD, G. L. C. 1917 Economic significance of the root development of agricultural crops. *Agr. Jour. India, Indian Sci. Cong. No.* 1917: 17-28 (Abs. in *Exp. Sta. Rec.* 39: 230).
- (30) KIESSELBACH, T. A. 1910 Transpiration experiments with the corn plant. *Nebr. Agr. Exp. Sta. Ann. Rpt.* 24: 125-139.
- (31) KIESSELBACH, T. A., RUSSEL, J. C., AND ANDERSON, A. 1929 Significance of subsoil moisture in alfalfa production. *Jour. Amer. Soc. Agron.* 21: 241-268.
- (32) LEBEDEV, A. F. 1928 Volume weight of soils as a physical characteristic of the soil profile. *Soil Sci.* 25: 207-211.
- (33) McMILLER, P. R. 1919 Some notes on the cause of unproductivity of raw subsoils in humid regions. *Soil Sci.* 7: 233.
- (34) METZGER, W. H., AND GRANDFIELD, C. O. 1938 Extension of alfalfa roots into subsoil dried by a previous crop. *Jour. Amer. Soc. Agron.* 30: 80.
- (35) MILLAR, C. E. 1925 Availability of nutrients in subsoils. *Soil Sci.* 19: 275.
- (36) MILLAR, C. E. 1933 Availability to corn of nutrients in the A₂ and B horizons of Hillsdale loam. *Jour. Amer. Soc. Agron.* 25: 418-426.
- (37) MILLER, E. C. 1916 Root systems of agricultural plants. *Jour. Amer. Soc. Agron.* 8: 129-154.
- (38) MORGAN, M. F. 1937 Universal soil testing system. *Conn. Agr. Exp. Sta. Bul.* 392.
- (39) NISSLEY, C. H. 1939 Vegetable crops respond to deep tillage. *N. J. Agr.* 21 (5): 3-4.
- (40) PRINCE, A. L., TOTHE, S. J., AND BLAIR, A. W. 1938 Chemical composition of soil from cultivated land and from land abandoned to grass and weeds. *Soil Sci.* 46: 379-389.
- (41) SEANTZ, H. L. 1925 Soil moisture in relation to the growth of crop plants. *Jour. Amer. Soc. Agron.* 17: 705-711.
- (42) TIEDJENS, V. A. 1939 Improving subsoils for vegetable production. *N. J. State Hort. Soc. News* 20: 1091-1092.
- (43) THORNTON, S. F. 1935 Available phosphorus and potassium contents of surface soils and subsoils as shown by the Neubauer method and by chemical tests. *Jour. Amer. Soc. Agron.* 27: 46-51.
- (44) VAN DER MERWE, C. R. 1926 Contribution to the study of the infertility of the subsoil. *Jour. Agr. Sci.* 16: 507-520.
- (45) WASHEKO, J. B. 1938 Factors affecting yielding ability of four varieties of soybeans. Master's thesis, Rutgers University, New Brunswick, N. J.
- (46) WATENPAUGH, H. N. 1936 Influence of the reaction of soil strata upon the root development of alfalfa. *Soil Sci.* 41: 449-468.
- (47) WEAVER, J. E. 1926 Root Development of Field Crops. McGraw-Hill Book Co., New York.
- (48) WEAVER, J. E., AND CRIST, J. W. 1922 Relation of hard-pan to root penetration in the Great Plains. *Ecology* 3: 237-249.
- (49) WEAVER, J. E., HOUGEN, V. H., AND WELDON, M. D. 1935 Relation of root distribution to organic matter in prairie soil. *Bot. Gaz.* 96: 389-420.
- (50) WEAVER, J. E., JEAN, F. C., AND CRIST, J. W. 1922 Development and activities of roots of crop plants. Carnegie Inst. Wash. Pub. 316.

SORPTION BY CLAYS

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The uptake of water vapor by soils and clays has been studied frequently, either to elucidate the mechanism of the process, or to reveal relations between uptake and the physical properties or chemical composition of the clay. The present paper shows that these objectives are better attained if the soils and clays are exposed, not only to water vapor, but also to the vapor of an organic liquid such as toluene. A similar treatment was applied by Mitscherlich (21), who exposed soil to water vapor and to benzene vapor.

The study of the uptake of vapor by soils was started in 1814 by De Saussure (24), who noted evolution of heat when dry solids were moistened by various vapors. In the case of soil exposed to water vapor, the moisture content (per cent by weight) beyond which no heat is liberated is called the "hygroscopic coefficient." As so defined, the hygroscopic coefficient can be calculated from the heat of wetting (19, 26), but it is usually estimated (20, p. 69) by exposing dry soil over 10 per cent sulfuric acid until there is no further measurable uptake. Keen (12) has noted, however, that the hygroscopic coefficient is neither strictly defined nor capable of precise measurement.

In the face of the vagueness of the term "hygroscopic water," and the ignorance of the processes involved, the uptake of vapors by soils and clays is a process to which McBain's (18) term "sorption" may legitimately be applied.

Adsorption in unimolecular layers does not appear to be of first importance in this sorption, for Mattson (17), having estimated the total available surface presented by a soil colloid, has calculated that the hygroscopic moisture was sufficient to provide a layer 16 molecules thick. Ehrenberg (4), by a similar calculation, estimated that the layer of hygroscopic water could be 10 molecules deep.

Besides the concept of thick layers of liquid evolved by Mattson and Ehrenberg, a capillary condensation mechanism is also to be found in the literature. It seems probable that with either mechanism, the extent of the sorption would depend, not so much on the specific action of the solid on the vapor, as on the pressure and molecular volume of the vapor and the surface tension and compressibility of the liquid. Zunker (27) has discussed the relative sorptions of water and organic liquids from this point of view, holding that

¹ It is a pleasure to acknowledge the assistance and encouragement received during the course of this work from H. Greene, of this institute.

surface tension is the principal factor controlling sorption, and concluding that the sorption of aromatic hydrocarbons should, on this account, be smaller than that of water. He quotes Neugebohrn (23), who gave figures for sorption of benzene by a loam and a clay which are respectively 95 and 84 per cent of those for sorption of water. Mitscherlich (21) measured sorption of benzene by soils exposed over a mixture of benzene with olive oil. The figures multiplied by 4.4 were subtracted from the hygroscopic coefficients to give values for "intracellular" surface. This rested on the view that the hygroscopic coefficient measured the total surface, and the benzene figures, the outer surface of the soil material.

The subject has acquired a different aspect since Hendricks and Fry (9) and Kelley, Dore, and Brown (13) showed by x-ray measurements that clays contain definite clay minerals, of which the two principal ones are of the kaolinite and montmorillonite types. Kaolinite and montmorillonite are very different substances in their relations to water. From the vapor, kaolinite sorbs very much less water than does montmorillonite; and in the liquid, montmorillonite clays swell noticeably, whereas kaolinite clays do not. This difference between the two clay types was emphasised by the discovery by Hofmann, Endell, and Wilm (10) that the crystal lattice of montmorillonite shows a marked unidimensional shrinkage on dehydration. Kaolinite does not show this lattice change on dehydration.

Montmorillonite itself has this specific relation to water manifested in its reversible lattice swelling, which is not present with kaolinite and hydrous mica. All the clay minerals, however, hold water as lattice hydroxyl groups, and they may all have water associated with their exchangeable cations. Many clay minerals, as removed from soil, contain, besides clay mineral, free sesquioxides and admixed quartz, and water may be sorbed by these substances as well as by the clay minerals.

The way in which water is lost from soil colloids on heating has been studied by Kelley and co-workers (14) and by Hendricks and Alexander (8). The results of these dehydration studies have also shown how fundamentally different in their relations to water are kaolinite and montmorillonite clay types.

Nagelschmidt (22) in his study of the reversible lattice contraction of montmorillonite gives 12.3 Å. for the d_{001} of oven-dry montmorillonite containing 9.8 per cent water, and 15.2 Å. for the same material containing 23.4 per cent water. From his curve it appears that of the 13.6 per cent difference in moisture content about 3 units are accommodated between the sheets of the montmorillonite structure and about 11 units are reversibly taken up in other ways. Nagelschmidt also used methylene iodide and methyl iodide instead of water; he was unable to observe any lattice swelling, but the amounts taken up were probably too small to show the effect even if the whole of the liquid had penetrated between the sheets of the lattice.

Barrer (2) working on the sorption of polar and nonpolar gases by zeolites

has shown that comparison of the sorption properties for water and ammonia and for the permanent gases can be used to determine whether the zeolite lattice is three-dimensional, fibrous, or platy. Chabasite and analcite, zeolites with rigid three-dimensional lattices which do not collapse on dehydration, take up at saturation similar quantities of water vapor and inert gases. Heulandite is a zeolite of a different type; the lattice is not rigid, but is a laminated structure with interlaminal water, some of which is very loosely bound. Dehydration is accompanied by lattice shrinkage, and, though ammonia is sorbed by the dehydrated lattice, the inert gases are taken up in insignificant quantities.

The purpose of this discussion has been to show that clays will, in general, sorb water in what may be regarded as a dual manner. As they are porous solids of large surface area, they will sorb water vapor, or indeed any condensable vapor, by condensation processes. They can also take up water vapor by forces which are specific to water, for example, as water of lattice swelling, in the particular case of montmorillonite, as water associated with exchangeable bases, or as water of hydration of admixed sesquioxides.

In the present paper are reported measurements of sorption of water vapor and of toluene vapor by some clays separated from Sudan topsoils (1-foot layer) by sedimentation in water. The clays are selected from those described by Greene (7), who gives analyses and pedological data. The differences, water sorption minus toluene sorption, are shown to be related to the silica-sesquioxide ratios of the clays. If it were assumed that the same processes were at work in sorption of water and toluene it would be natural to consider the ratios of one to the other and to explain these ratios as resulting from differences in compressibility, vapor pressure, surface tension, and the like. It will be appreciated, however, in the light of the foregoing discussion, that water may be sorbed partly as toluene is sorbed, but in addition water may be sorbed in ways, or in situations, which are not available to the organic vapor. For this reason it is appropriate to consider differences rather than ratios. A similar point of view was adopted by Winterkorn and Baver (25), who compared the unrestricted uptake of liquid water and certain organic liquids by clay colloids. They used the uptake of benzene as a measure of the pore space, and the difference between this and the water uptake as the water concerned with swelling. They found the xylene and toluene uptakes were approximately the same for all the colloids used, whereas uptakes of water showed considerable differences. Similarly, the determination of "xylene equivalents" by the use of xylene instead of water in the standard determination of the moisture equivalent was suggested by Fisher (5). Joseph (11) made a number of determinations of this quantity and of the difference between it and the moisture equivalent. Values of "imbibitional water" given by Fisher show clearly the additional information obtained by this means. It is interesting to note that at this higher level of moisture content sodium clay was found by Joseph to hold much more imbibitional water than calcium clay, a difference

which is not appreciable at low moisture contents. For example, the work of Anderson (1) and others has shown that, of the common cations, only potassium has a marked effect on hygroscopicity over 30 per cent sulfuric acid, that is, at low moisture contents. Potassium is usually a minor constituent of the replaceable bases in natural clays, and these are not therefore likely to show cationic effects in sorption measurements.

EXPERIMENTAL

The difference between the sorption specific to water and that which operates with any condensable vapor may be illustrated by the following results obtained with soil and charcoal to which sodium chloride had been added.

Two 5-gm. lots of finely divided wood charcoal were each treated with 10 cc. of 0.5 *N* NaCl solution, and two other samples of charcoal were treated with 10 cc. of distilled water. The four samples were oven dried, and one of each

TABLE 1
Sorption of toluene and of water by charcoal
Percentage oven-dry weight

TIME OF EXPOSURE.....days	1	2	3	4	5	7
<i>Water sorption</i>						
Salt-treated.....	5.8	8.8	12.2	16.2	19.8	25.6
Water-treated.....	6.1	7.9	7.9	8.1	8.1	8.4
<i>Toluene sorption</i>						
Salt-treated.....	3.0	4.3	5.0	5.6	6.1	6.8
Water-treated.....	4.5	6.1	6.9	7.5	8.0	8.75

treatment was then exposed to water and one to toluene vapor. The progress of the sorptions was followed by daily weighing.

The addition of sodium chloride is intended to endow one pair of samples with a sorption specific to water. The figures in table 1 show that the salt-treated sample has a large sorption of water; but the sorption of toluene is not very different from that of the sample treated merely with water. Furthermore, the two samples not treated with salt show similar sorptions of toluene and water.

Table 2 shows similar results from the use of Gezira topsoil instead of charcoal. Here again, addition of a specific water attractant affects the uptake of toluene but little, though it has a large effect on the sorption of water. The fact that the water sorption of this soil, even without salt, is considerably greater than the toluene sorption, confirms the belief that Gezira soil has a specific attraction for water. This is in accord with the fact that the clay of the Sudan Gezira has a high silica-sesquioxide ratio and swells considerably in water.

The curves in figure 1 representing the sorption of water and of toluene by Chalamni and Yirrol Mound, two clays of widely different silica-sesquioxide ratios, illustrate the nature of the process. For simplicity of operation, these and the other samples were exposed in desiccators containing standardized volumes of pure water or toluene, the surface area being the same for the two

TABLE 2
Sorption of toluene and of water by Gezira soil
Percentage oven-dry weight

TIME OF EXPOSURE.....days	1	2	3	4	5	7
<i>Water sorption</i>						
Salt-treated.....	7.2	11.8	14.9	17.8	21.0	25.8
Water-treated.....	7.9	10.2	10.7	10.9	11.0	11.5
<i>Toluene sorption</i>						
Salt-treated.....	3.6	3.1	4.4	4.5	4.6	5.0
Water-treated.....	4.4	4.9	5.1	5.3	5.5	5.8

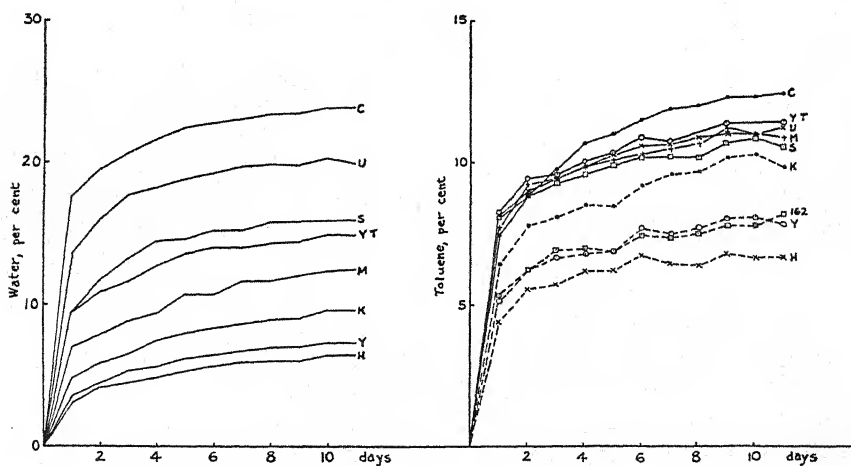


FIG. 1. SORPTION OF WATER AND OF TOLUENE BY SUDAN CLAYS

Note difference in vertical scales, expressing weight of sorbed liquid as percentage of oven-dry weight of clay. C = Chalamni, U = Umm Berembeita; S = Sue; YT = Yirrol Toich; M = Mboro; K = Kagelu; 162 = Site 162; Y = Yirrol Mound, H = Halima.

liquids. Under these conditions, equilibrium values were not expected, and, as can be seen from the curves, no close approach to equilibrium was achieved in the 11-day period of observation. Although, by using solutions of sulfuric acid in water and of toluene in a nonvolatile oil, equilibrium sorptions could doubtless have been reached, it seemed likely that any specific sorption of water

would be no less readily observed in the earlier stages of uptake over the pure liquid. The results depend on the total quantity of material exposed in the desiccator because, in the early stages, sorption is rapid enough to reduce the vapor pressure above the clay well below the saturation value. Care was taken, therefore, to use the same number of samples of the same weight in the same-sized containers in each series of measurements. The desiccator was thermostatically controlled at 23°C. It should be noted that the same clay samples were used for the sorption of both liquids. This has certain disadvantages; on the other hand, it brings into relief the changes brought about in clays during oven drying, which are referred to later. As is well known, one of the most troublesome matters in work of this kind is the degree of preliminary drying to which the samples are subjected. In the present work this was not specially studied, the clays being simply left for 20 hours in an oven running at 105°C.

Results obtained for daily sorption of water and of toluene for other clays during an 11-day period are also shown in figure 1. Considerable similarity will be noted between the two sets of curves. In both, the highly siliceous clay Chalamni, which according to unpublished observations of Nagelschmidt contains much montmorillonite, is at the top, closely followed by Umm Berembeita, which is of similar character. Both are gray alkaline clays of depressions, moistened for only a part of each year, during the rainy season, after which they dry and crack deeply. The four lowest curves are for clays of low silica content probably containing kaolinite or halloysite but no montmorillonite; of the three clays that have an intermediate position in both diagrams, two, Sue and Yirrol Toich, are somewhat like Chalamni and Umm Berembeita in their topographical situation but differ from them, it is thought, in being formed from material which was previously leached.

From this rather remarkable relation of the toluene sorption to the silica content of the clays, it may be concluded that clays containing much silica have a physical structure favorable to the condensation even of organic vapors. These are the clays which swell in water and to a notable degree also in sodium carbonate solution and which probably contain much montmorillonite. It is possible, therefore, that swelling and ability to sorb a higher proportion of organic liquids are both consequences of the montmorillonite structure.

Table 3 shows successive daily values of the quantity, water uptake minus toluene uptake. It will be seen that these figures clearly differentiate the various clays. Initial values for the two alkaline clays Chalamni and Umm Berembeita are 10.2 and 5.6. Next in order are Sue and Yirrol Toich, clays of lower silica content but having a somewhat similar topographical situation. It is a feature of these clays, accumulated in low situations, to be enriched to a greater or less extent with material leached from higher ground, that is, with silica and bases. The remaining five clays, which are of low silica content, do not share this characteristic and are here seen to sorb slightly more toluene than water. It is not surprising that in such a group of representative clays

TABLE 3
Differences between water and toluene sorptions by Sudan clays
 Water sorption less toluene sorption, percentage oven-dry weight

	SITE	SiO ₂ R ₂ O ₃	SiO ₂ Al ₂ O ₃	1 DAY	2 DAYS	3 DAYS	4 DAYS	5 DAYS	6 DAYS	7 DAYS	8 DAYS	9 DAYS	10 DAYS	11 DAYS
Chalanni.....	127	3.56	4.43	10.2	10.6	11.0	10.9	11.4	11.2	11.2	11.3	11.1	11.4	11.4
Umm Berembelta.....	Nuba Mts.	2.83	3.77	5.6	7.0	8.3	8.3	8.6	8.6	9.0	9.0	8.6	9.2	8.7
Sue.....	179	2.23	2.57	1.3	2.8	4.0	4.8	4.7	4.9	5.0	5.5	5.0	4.9	5.2
Yirrol Toich.....	204	2.13	2.50	1.2	1.5	2.0	2.7	3.2	3.0	3.3	3.2	3.0	3.4	3.4
Halima.....	189	2.18	2.46	-1.2	-1.4	-1.2	-1.3	-0.9	-0.9	-0.6	-0.6	-0.8	-0.3	-0.4
Mboro.....	183	1.75	2.22	-0.6	-1.3	-0.5	-0.5	+0.6	+0.4	+0.9	+0.9	+0.8	1.3	1.3
Site 162.....	...	1.77	2.16	-1.5	-1.8	-1.6	-1.3	-0.8	-1.3	-0.9	-0.8	-1.2	-0.8	-0.5
Kagelu.....	52	1.92	2.12	-1.5	-1.9	-1.6	-1.0	-0.5	-0.9	-1.0	-0.8	-1.2	-0.8	-0.4
Yirrol Mound.....	212	1.84	2.07	-1.7	-1.6	-1.5	-1.2	-0.7	-1.1	-0.7	-0.5	-0.7	-0.6	-0.9

there should be no sharp division into two classes without intermediate members. The extreme members show wide differences in behavior, which probably can be associated with the montmorillonite type and with the kaolinite type. The question as to whether the intermediate clays are mixtures of clay minerals, mixtures of clay minerals with other minerals and oxides, or clay minerals with mixed lattices, cannot be settled on the results of this investigation alone.

Investigations, particularly on American soils, have shown how widespread is this admixture in soil colloids. For example, Kelley and co-workers (15, 16) have shown that Susquehanna colloid contains both kaolinite and montmorillonite in some horizons and that all the San Joaquin colloids contain a third clay mineral and kaolinite, and Nagelschmidt (22) has found kaolinite lines in the x-ray spectrum of montmorillonite from Montmorillon. The presence of free sesquioxides of iron and aluminum, and of free silica has frequently been reported in soil colloids.

TABLE 4
Differences by volume, water sorption minus toluene sorption by Sudan clays

CLAY	SiO ₂ /R ₂ O ₃	1 DAY	11 DAYS
Chalamni.....	3.56	9.0	9.3
Umm Berembeita.....	2.83	4.3	6.9
Sue.....	2.23	0.0	3.5
Yirrol Toich.....	2.13	-0.1	1.5
Halima.....	2.18	-1.9	-1.5
Mboro.....	1.75	-1.8	-0.3
Site 162.....	1.77	-2.3	-1.9
Kagelu.....	1.92	-2.5	-2.1
Yirrol Mound.....	1.84	-2.6	-2.2

It should be emphasized that in the present paper what is measured is the weight of water or toluene sorbed, and the figures recorded are percentages by weight. They cannot be expressed as volumes without making assumptions as to the degree of compression existing in the sorbed liquids and thus introducing an unnecessary complication. As a matter of interest, however, table 4 shows 1-day and 11-day values for differences by volume (cubic centimeters per 100 gm. soil) assuming densities of sorbed water and toluene to be respectively 1.00 and 0.86.

It is clear that on either basis the differences in sorption are fairly closely related to the silica content and pedological character of the clay and that this method of investigation is more informative than simply measuring hygroscopicity. The specimens examined were extracted clays. When soils are studied there is, of course, the additional reason for using an organic liquid as well as water, that the results will primarily depend on the amount of clay present, since the latter largely defines the total surface and mean capillary size of the material under examination.

Under special conditions, hygroscopicity has been used as a measure of clay content. Thus, Gile and co-workers (6) found 95 per cent of the water uptake to be due to the clay. These authors found that hygroscopicity is more closely dependent on the silica-sesquioxide ratio when it is measured over strong sulfuric acid than when measured over 3 per cent acid. A similar observation was made by Bayer and Horner (3). It appears from the figures of the latter workers that the relation was particularly evident for colloids having low silica-sesquioxide ratios. Apparently, therefore, the effect of silica content on sorption is most evident at low moisture contents. To some extent, this conclusion contrasts with the results now recorded, in which the differences appear equally evident after exposures of 1 day and of 11 days. This suggests that specific sorption of water by clay is a notably speedy process which is overlaid by a nonspecific sorption of a rather uniform character, and that, consequently, differences established at first remain unobscured. From the practical

TABLE 5
Water sorption minus toluene sorption by Sudan clays after 9 days
Percentage oven-dry weight

	SERIES 1	SERIES 2	SERIES 3
Chalamni.....	14.1	12.2	11.1
Umm Berebeita.....	10.8	8.3	8.6
Suc.....	8.2	6.2	5.0
Yirrol Toich.....	5.3	5.2	3.0
Halima.....	0.7	0.7	-0.8
Mboro.....	2.3	2.3	+0.8
Site 162.....	0.5	1.3	-1.2
Kagelu.....	1.5	2.1	-1.2
Yirrol.....	0.3	1.5	-0.7

point of view, it is satisfactory that a short period of exposure suffices to bring out characteristic differences.

Table 5 records figures obtained in three successive series of measurements on these clays. Undoubtedly part of the discrepancy in these results is due to variations in the extent of drying, but the variations between the series are not always in the same direction, and it seems probable that changes taking place on oven drying are partly responsible.

SUMMARY

McBain's term "sorption" is applied to the whole process of vapor uptake by soils and clays. Earlier work on the sorption of vapors is discussed, and the importance of the fundamental differences between the behavior toward water of montmorillonite and kaolinite clay types is stressed.

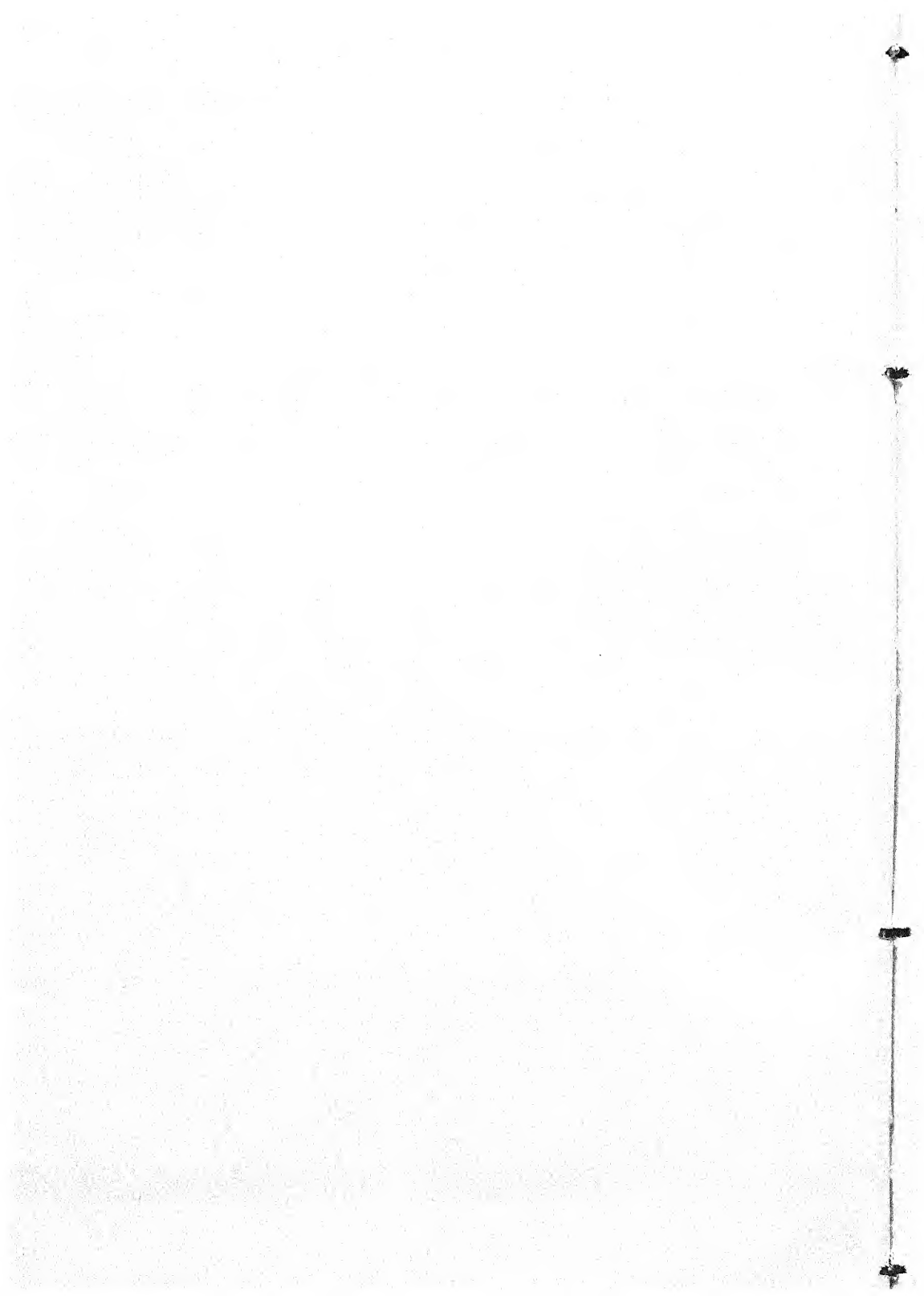
It is considered that the basis of this difference lies in differences in the specific attraction for water of the two minerals. Toluene sorptions of a

number of Sudan clays have been measured. The sorption of toluene is regarded as a particular instance of the sorption which takes place with any condensable vapor, and which is a measure of such quantities as surface area and capillary distribution. The water sorptions of the same clays have been measured; though water is sorbed physically by the same mechanism as toluene, it can also be held by forces which are specific to water. It has been found that soil colloids with low silica contents sorb, under the conditions of these experiments, similar amounts of water and toluene; those with high silica-sesquioxide ratios sorb much more water than toluene. These differences are also related to the pedological nature of the clays.

REFERENCES

- (1) ANDERSON, M. S. 1927 Influence of substituted cations on the properties of soil colloids. *Jour. Agr. Res.* 38: 565-584.
- (2) BARRER, R. M. 1938 Sorption of polar and nonpolar gases by zeolites. *Proc. Roy. Soc. [London]* (A) 167: 392-420.
- (3) BAYER, L. D., AND HORNER, G. M. 1933 Water content of soil colloids as related to their chemical composition. *Soil Sci.* 36: 329-353.
- (4) EHRENBERG, P. 1922 *Die Boden Kolloid*, ed. 3. Dresden and Leipzig.
- (5) FISHER, E. A. 1924 Remarks and observations on imbibitional soil moisture. *Jour. Agr. Sci.* 14: 204-220.
- (6) GILE, P. L., ET AL. 1924 Estimation of colloidal material in soils by adsorption. U. S. Dept. Agr. Bul. 1193.
- (7) GREENE, H. 1939 Einige Böden der Äquatorial-Provinz des anglo-ägyptischen Sudans. *Soil Res.* 6: 325-338.
- (8) HENDRICKS, S. B., AND ALEXANDER, L. T. 1939 Minerals present in soil colloids: I. *Soil Sci.* 48: 257-268.
- (9) HENDRICKS, S. B., AND FRY, W. H. 1930 Results of x-ray examinations of soil colloids. *Soil Sci.* 29: 457-476.
- (10) HOFMANN, U., ENDELL, K., AND WILM, D. 1933 Kristallstruktur und Quellung von Montmorillonit. *Ztschr. Krist.* 86: 340-348.
- (11) JOSEPH, A. F. 1924 Report of government chemist. Wellcome Trop. Res. Lab., Khartum. [See also Fisher (5).]
- (12) KEEN, B. A. 1931 The Physical Properties of the Soil. Longmans Green. [See also, PURI, A. N., CROWTHER, E. M., AND KEEN, B. A. 1925 Relation between the vapour pressure and water content of soils. *Jour. Agr. Sci.* 15: 68-88.]
- (13) KELLEY, W. P., DORE, W. H., AND BROWN, S. M. 1931 Nature of the base-exchange material of bentonite, soils, and zeolites, as revealed by chemical investigation and x-ray analysis. *Soil Sci.* 31: 25-45.
- (14) KELLEY, W. P., JENNY, H., AND BROWN, S. M. 1936 The hydration of minerals and soil colloids in relation to crystal structure. *Soil Sci.* 41: 259-274.
- (15) KELLEY, W. P., ET AL. 1939 Colloidal constituents of California soils. *Soil Sci.* 48: 201-255.
- (16) KELLEY, W. P., ET AL. 1939 Comparative study of the colloids of a Cecil and a Susquehanna soil profile. *Soil Sci.* 47: 175-193.
- (17) MATTSON, S. 1932 Laws of soil colloidal behavior: VIII. Forms and functions of water. *Soil Sci.* 33: 301-322.
- (18) MCBAIN, J. W. 1931 Sorption of Gases and Vapours by Solids. London.
- (19) MIRTSCH, H. 1930 Eine Bestimmung der Benetzungswärme des Bodens. *Bot. Arch.* 28: 451.

- (20) MITSCHERLICH, E. A. 1923 *Bodenkunde für Land und Forstwirte*, ed. 4. Parey, Berlin.
- (21) MITSCHERLICH, E. A. 1923 *Abdehalden's Handbuch der Biologischen Arbeits Methoden* 11: 276.
- (22) NAGELSCHMIDT, G. 1936 On the lattice shrinkage and structure of montmorillonite. *Ztschr. Krist.* 93: 481-487.
- (23) NEUGEBOHRN, A. 1927 Die Bestimmung der Bodenoberfläche durch Flüssigkeitsabsorption. *Kulturtechniker* 30: 242.
- (24) SAUSSURE, TH. DE 1814 Beobachtung über die Absorption der Gasarten durch verschiedene Körper. *Gillb. Ann.* 47: 113.
- (25) WINTERKORN, H., AND BAVER, L. D. 1934 Sorption of liquids by soil colloids: I. Liquid intake and swelling by soil colloidal materials. *Soil Sci.* 38: 291-298.
- (26) ZUNKER, F. 1928 Über das Schwinden und Quellen der Böden und ein neues Bodenuntersuchungsverfahren. *Kulturtechniker* 31: 527.
- (27) ZUNKER, F. 1930 Das Verhalten des Bodens zum Wasser. *Blanck's Handbuch der Bodenlehre* 6: 78.



EFFECT OF VARIOUS SOIL TREATMENTS ON NITRATES, SOIL MOISTURE, AND YIELD OF WINTER WHEAT¹

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In the early development of dry farming, many data were accumulated which showed the advantage of early plowing and subsequent thorough seed-bed preparation in the production of winter wheat. The improved yields from these practices were generally attributed to the moisture conserved. One of the first studies which showed that moisture was only one of the contributing factors to yield was reported by Buckman (2), who found that wheat yields on the dry lands of Montana were closely correlated with the amount of nitrates in the soil. Somewhat later, Call (3), of Kansas, likewise concluded that the supply of available nitrogen was an important factor in the production of dry-land grain. Several similar studies have also been reported showing the relationship of nitrate accumulation to crop production.

The purpose of this study was to determine the relationships of yield of winter wheat under dry-farm conditions to amounts of available nitrogen and of soil moisture as influenced by time of plowing and manurial treatments. Most of the data reported were obtained between 1928 and 1933, inclusive.

EXPERIMENTAL METHODS

The experiment was conducted at the Nephi Dry-Land Station in central Utah. The soil of the area, derived from the weatherings of the adjacent mountain range, reddish-brown, and uniformly 10 or more feet deep, is classed as a clay loam. At the moisture equivalent point the soil was found to retain approximately 22 per cent moisture. The mean total nitrogen content of the plots at the beginning of the test was found to be as follows: 1 foot, 0.1043 per cent; 2-3 feet, 0.0727 per cent; 3-6 feet, 0.0434 per cent.

For nitric nitrogen and soil moisture determinations, six samples taken from each plot with a King soil tube, to the depths and on the dates later indicated, were carefully mixed. All determinations were made in duplicate. Moisture contents are based on the weight of soils dried to 105°C. Nitric nitrogen is expressed on the basis of air-dry soil and was determined by the usual phenol-

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disulfonic acid method. All results involving soluble nitrogen are expressed as nitric nitrogen (N) or elemental nitrogen in the NO_3 form.

The cropping system consisted of alternate winter wheat and fallow. The various soil treatments were made during the fallow year. The plots used in the time of plowing tests were $\frac{1}{10}$ acre in size, and those for the barnyard manure treatment were $\frac{1}{20}$ acre. Each test was duplicated.

RESULTS AND DISCUSSION

Weather data

Since moisture and temperature are two of the controlling factors governing accumulation of available nitrogen and, in turn, crop yields, the monthly mean temperatures (table 1) and the monthly rainfall (table 2) for April to October, 1928 to 1933, inclusive, are given.

TABLE 1

Mean monthly temperatures, in degrees F., April to October, 1928 to 1933, inclusive, together with means for the period 1908 to 1923

YEAR	TEMPERATURE						
	April	May	June	July	Aug.	Sept.	Oct.
1928	43.2	58.0	61.2	72.0	69.0	62.0	50.0
1929	43.1	53.0	62.1	72.3	71.0	58.0	47.0
1930	51.6	51.2	63.3	72.7	69.4	59.4	44.7
1931	45.4	53.0	67.4	75.0	71.5	60.9	52.0
1932	49.0	54.4	61.6	71.0	70.0	61.0	45.3
1933	42.2	47.2	67.8	76.3	70.1	64.4	54.2
Mean.....	45.7	52.8	63.9	73.2	70.2	60.9	48.9
Mean 1908-1923...	44.0	53.0	63.0	71.0	70.0	61.0	48.0

The mean temperatures over the period of this experiment differed little from the averages since 1908, with the exception of July. The long-time mean for July was 71, as compared with 73.2 for the period from 1928 to 1933, which made conditions more critical for wheat growth particularly in the latter stages of development.

With the exception of 1929 and 1930 the rainfall from 1928 to 1933 was below the long time normal of 13.21 inches. In these two years the 4.97 inches for July and early August, 1929, and the 5.05 inches for August and September, 1930, fell in such heavy torrential showers that a large part of the moisture was lost through runoff. According to Bracken and Cardon (1), not only is all the summer rainfall at the Nephi Station lost primarily by evaporation but an average 10.5 per cent of that remaining in the soil from the winter precipitation is also lost. Over the whole fallow crop cycle including two winters and a summer they found that only 32.5 per cent of the rainfall was stored as soil moisture.

Yield, nitric nitrogen, and soil moisture as affected by time of plowing and by various amounts of barnyard manure

Time of plowing, aside from weed control following plowing, is recognized as the most important single tillage factor contributing to the successful production of crops on the dry farms of Utah. This is indicated by the data in table 3 showing yields of winter wheat for the years 1928 to 1933, inclusive, for fall plowing and plowing at various delayed periods in the spring. A difference of only 0.9 bushel is shown between fall plowing and plowing delayed in spring until the volunteer was 6 inches high. According to the variance analysis of the data, this difference is beyond the 1 per cent point, but an examination of the results for the individual years shows essentially no difference except in 1929 when fall-plowed plots produced 18.3 bushels an acre and spring plowing

TABLE 2

Monthly rainfall, in inches, 1928 to 1933, inclusive, together with totals for the respective years, monthly means from 1928 to 1933 inclusive, and means for the period 1898 to 1933

YEAR	RAINFALL												
	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.	Total
1928	0.41	0.68	1.56	0.77	1.52	0.41	0.79	0.20	0.16	0.84	1.64	0.59	9.57
1929	0.79	0.91	1.44	1.22	0.85	0.85	3.46	1.80	0.85	0.81	0.26	0.45	13.69
1930	1.77	1.12	2.22	0.88	2.58	0.17	1.44	2.44	2.61	1.41	0.72	0.18	17.54
1931	0.14	0.40	0.81	0.96	0.74	0.03	0.32	1.08	0.29	0.47	1.17	1.01	7.42
1932	1.14	1.50	1.90	0.80	0.52	1.09	0.59	1.14	0.03	1.05	0.26	0.75	10.77
1933	1.10	0.15	1.07	1.21	2.79	0.17	0.80	0.55	0.19	0.28	0.30	0.72	9.33
Mean 1928-1933.....	0.89	0.79	1.50	0.97	1.50	0.45	1.18	1.20	0.69	0.81	0.72	0.62	11.32
Mean 1898-1933.....	1.16	1.10	1.40	1.31	1.47	0.60	0.95	1.18	1.01	1.13	0.90	1.00	13.21

when the volunteer was 6 inches high produced 11.7 bushels. If this difference, which might have been the result of some unusual seasonal condition, is discarded, the conclusion might be drawn that 18.9 bushels is not significantly different from 18.0 bushels. The still later plowings for fallow show much greater reductions in yield. That early cultivation not only has a distinct advantage in respect to wheat yields in Utah and generally on the dry lands of the far west, as reported in a number of publications, but likewise on the Great Plains, is indicated by results reported by Sewell and Gainey (8, 9). These workers found over a 20-year period in Kansas that July plowing, which is considered early in that area, produced an average increase of 10 bushels of winter wheat per acre over September plowing.

Both the time of plowing and the barnyard manure tests were started in 1916. From the beginning of these tests until 1933 the averages for fall

plowing in the time of plowing test and those for spring plowing with no treatment in the barnyard manure test were approximately equal, but during the cycle of low rainfall, the early spring plowing given the plots in the barnyard manure test produced an average yield of 21.3 bushels as compared with only 18.9 bushels for fall plowing in the time of plowing test. The statistical analysis of the results in the barnyard manure test showed that the ratio of the error figure to the mean square for treatments reached only the 5 per cent level of significance. If 2.14 bushels is used as a basis of comparison

TABLE 3

Acre yields, in bushels, of Utah Kanred winter wheat produced on plots plowed at various times of the season for fallow and on plots treated with different amounts of barnyard manure, 1928 to 1933, inclusive

TREATMENT	WHEAT YIELDS						
	1928	1929	1930	1931	1932	1933	Mean
<i>Time of plowing</i>							
Fall plowed.....	20.9	18.3	20.1	22.5	17.5	13.8	18.9
Spring plowed, volunteer 6 inches high.....	21.1	11.7	20.7	22.1	17.8	14.6	18.0
Spring plowed, volunteer 12 inches high.....	14.4	8.1	18.1	17.7	17.7	10.2	14.4
Spring plowed, volunteer in bloom...	8.4	8.1	18.0	17.7	16.1	9.1	12.9
Spring plowed, volunteer in dough...	5.1	9.0	15.4	14.7	13.7	8.4	11.1
Mean.....	14.0	11.0	18.5	18.9	16.6	11.2
Differences for comparison of treatment means							
{ 5 per cent point.....							.50
{ 1 per cent point.....							.68
<i>Manurial</i>							
Spring plowed fallow, no manure....	28.1	21.4	20.1	23.5	15.8	19.0	21.3
Spring plowed fallow, 5 tons manure each alternate year.....	33.2	25.2	24.2	25.0	22.0	19.5	24.8
Spring plowed fallow, 10 tons manure each alternate year.....	35.1	25.0	21.5	21.6	25.8	19.3	24.7
Mean.....	32.1	23.9	21.9	23.4	21.2	19.3
Difference for comparison of treatment means, 5 per cent point.....							2.14

between treatment means, it is obvious that 21.3 bushels for no manure is significantly different from 24.8 bushels produced from the application of 5 tons of barnyard manure. Plots treated with 5 tons of manure and those to which 10 tons were applied showed no difference. In the earlier period of the test, however, the heavier treatment had an advantage, but crops produced on these plots in later years have "burned" in seasons of low rainfall. In the statistical analysis, the yearly means for both the time of plowing and the barnyard manure tests showed highly significant differences, or odds greater than 100 to 1 that

TABLE 4

Average soil moisture and nitric nitrogen contents of plots plowed at various times, 1928 to 1933

TIME OF FLOWING	CROPPED OR FALLOW	DEPTH	APRIL 15-20		JUNE 1-5		AUGUST 20-25		OCTOBER 20-25	
			Mois- ture	N	Mois- ture	N	Mois- ture	N	Mois- ture	N
		<i>feet</i>	<i>per cent</i>	<i>p.p.m.</i>	<i>per cent</i>	<i>p.p.m.</i>	<i>per cent</i>	<i>p.p.m.</i>	<i>per cent</i>	<i>p.p.m.</i>
Fall plowed for fallow	cropped	1	17.0	2.46	10.2	0.56	10.2	0.98	9.3	1.77
		2-3	18.2	3.11	11.5	0.50	11.4	0.51	10.4	1.04
		3-6	15.2	1.85	9.6	1.24
	fallow	1	16.8	1.41	15.1	1.94	13.1	2.81	13.7	4.78
		2-3	16.0	1.36	16.7	1.30	15.7	1.60	16.9	2.53
		3-6	11.7	1.19	13.8	1.65
Spring plowed, volunteer 6 inches high	cropped	1	17.6	2.01	11.0	0.65	9.8	1.19	11.3	1.74
		2-3	17.6	2.37	11.6	0.58	10.6	0.96	10.9	1.26
		3-6	15.6	1.75	10.1	1.09
	fallow	1	17.9	1.49	15.4	1.29	14.4	2.07	13.0	3.26
		2-3	16.5	1.42	15.9	1.27	15.6	1.53	15.1	2.56
		3-6	11.9	1.44	12.9	1.51
Spring plowed, volunteer 12 inches high	cropped	1	17.1	1.85	10.4	0.69	10.1	1.29	10.6	1.56
		2-3	17.1	2.38	12.2	0.46	10.8	0.56	10.4	1.01
		3-6	12.9	1.62	9.9	1.02
	fallow	1	16.8	1.37	11.6	0.93	11.2	1.74	12.1	2.48
		2-3	15.8	1.39	13.7	0.59	13.3	0.97	14.3	1.95
		3-6	11.3	1.23	11.2	1.75
Spring plowed, volunteer in bloom	cropped	1	17.0	1.57	9.5	0.27	10.2	0.84	11.2	1.49
		2-3	16.8	2.10	11.2	0.46	10.3	0.45	10.5	0.94
		3-6	12.7	1.51	10.3	1.05
	fallow	1	17.9	1.36	10.4	0.78	10.2	1.14	11.0	2.11
		2-3	15.8	1.63	12.4	0.50	11.3	0.58	11.7	1.81
		3-6	11.4	1.53	10.7	1.21
Spring plowed, volunteer in dough	cropped	1	17.2	1.54	10.6	0.40	10.3	1.14	10.2	1.87
		2-3	16.8	1.66	11.7	0.29	11.0	0.51	10.8	0.91
		3-6	12.8	1.54	9.9	1.13
	fallow	1	18.6	1.33	11.3	0.64	10.6	1.30	11.9	2.04
		2-3	16.5	1.42	12.5	0.61	11.7	0.71	12.5	1.44
		3-6	11.7	1.31	10.9	1.24

the differences were not the result of chance. Likewise, both showed significant interactions between years and treatments, indicating that the crop did not respond to the treatments in the same manner each season.

The explanation for the differences in yield shown in table 3 is the variation in accumulation of nitric nitrogen. The data are given in tables 4 and 5.

The data in table 4 show that there was a definite decrease in moisture percentage and amount of nitric nitrogen in fallow land in October with lateness of spring plowing, which bears a close relationship to acre yields as given in table 3. These data conform with results obtained by Sievers (10), who found

TABLE 5

Average soil moisture and nitric nitrogen contents of plots treated with various amounts of barnyard manure, 1928 to 1933, inclusive

MANURIAL TREATMENT	CROPPED OR FALLOW	DEPTH	APRIL 15-20		JUNE 1-5		AUGUST 20-25		OCTOBER 20-25	
			Mois- ture	N	Mois- ture	N	Mois- ture	N	Mois- ture	N
Spring plowed for fallow, no manure	cropped	feet	per cent	p.p.m.	per cent	p.p.m.	per cent	p.p.m.	per cent	p.p.m.
		1	17.0	2.19	10.6	0.71	10.3	1.12	10.8	1.56
		2-3	18.9	3.22	10.7	0.59	10.9	0.58	11.0	1.04
		3-6	18.6	2.02	11.6	1.11
	fallow	1	18.2	1.10	15.4	1.76	13.2	2.41	13.4	3.83
		2-3	17.2	1.49	17.0	1.32	16.5	1.50	16.1	2.36
		3-6	14.5	1.33	15.2	1.54
Spring plowed for fallow, 5 tons manure each alternate year	cropped	1	17.5	2.59	10.5	0.60	10.1	1.66	11.0	2.00
		2-3	18.2	4.59	10.9	0.76	10.3	0.83	10.3	1.29
		3-6	17.2	2.67	10.3	1.36
	fallow	1	18.3	1.34	15.1	2.08	13.9	4.18	13.9	6.10
		2-3	17.4	1.94	17.1	1.57	16.6	2.10	16.1	3.09
		3-6	13.2	1.47	14.4	2.49
Spring plowed for fallow, 10 tons manure each alternate year	cropped	1	17.2	5.54	10.8	0.59	10.9	1.78	11.2	2.45
		2-3	18.4	5.18	11.6	0.72	10.9	1.08	10.6	1.44
		3-6	17.4	3.10	11.8	1.48
	fallow	1	18.1	1.82	15.6	2.37	13.8	4.77	13.9	8.11
		2-3	17.1	2.02	17.0	2.03	16.1	2.39	16.0	4.44
		3-6	13.3	1.30	14.2	2.34

that dry-farm soils of Washington showed the greatest accumulation of moisture and available nitrogen when plowing was done early in the season. Unpublished data from the Moro Oregon Station indicate the same relationship. Call (3) also found that early tillage on the Great Plains was favorable to increase of nitrates.

The data showing the effect of no manure, 5 tons, and 10 tons applied each year just before plowing (table 5) indicate a marked increase in accumulation of nitric nitrogen for the manured fallow plots at the end of the season in

October and no appreciable difference in moisture percentages. The increased wheat yields for this part of the test (table 3) can be explained on the basis of the greater amount of available nitrogen.

Other relationships indicated by the data of tables 4 and 5 may be mentioned: (a) The nitric nitrogen remaining in the cropped plots at the June sampling was reduced to considerably less than 1 p.p.m. and, in a good many instances, to only a trace; (b) following the harvesting of the crop a slight gain in nitric nitrogen was usually made, even though there was no change in soil moisture content. While the comparison is not direct, Sewell and Gaine (8) found active nitrification occurring even though the soil was held at a low moisture percentage.

Results procured in 1923 with a shorter interval between sampling dates indicate characteristic changes in nitric nitrogen content of cropped plots as well as fallow plots (table 6).

TABLE 6
Nitric nitrogen content of cropped and fallow land on different dates in 1923

TREATMENT	DEPTH	NITRIC NITROGEN						
		5/10	5/31	6/21	7/15	8/7	9/2	10/5
	<i>feet</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>
Wheat	1	1.68	0.55	T	T	0.62	0.77	1.07
	2-3	3.82	1.50	0.60	T	0.58	0.65	0.75
	3-6	1.53	0.88	0.66	1.27	1.10	0.84	0.96
Fallow	1	1.03	1.13	2.22	4.00	3.57	4.50	5.28
	2-3	1.01	1.22	1.34	1.67	1.78	1.55	1.75
	3-6	1.40	1.69	1.68	1.78	1.79	1.67	1.94

The data in tables 4 and 5 show that the accumulation of nitric nitrogen in the fallow plots generally increased with the season, particularly for fall plowing. Even though moisture percentages were higher in the spring, the major part of the available nitrogen was accumulated in the latter part of the summer. The results shown in table 6 for fallow check plots are typical of this. As to time of season at which most active nitrification occurs, Leather (6) determined the nitrates for 6 months, from April to September, and found the greatest accumulation in June and July. Jensen (4) found that nitrates reached a peak on June 12, followed by a decrease. Smith (12) concluded that July was the month of greatest accumulation, and other workers have observed two peaks, one in late spring and another in early fall.

It might also be observed that the data obtained from the fallow plots (tables 4, 5, and 6) indicate that nitric nitrogen was formed mostly in the surface foot of soil.

From the data given (tables 4 and 5), a comparison may be made between the amount of nitric nitrogen formed at the end of the fallow season and that

remaining the following spring at the beginning of the crop year. Little difference is observed from such a comparison except that some movement to lower depths occurred, a result which is to be expected. A direct comparison in pounds per acre between the amount of nitric nitrogen found in fall and spring is given in table 7.

From the data shown in table 7 it is obvious that little change in amounts of nitric nitrogen occurred between fall and spring. Throughout the 5-year period over which this study was conducted, however, winter precipitation was not heavy enough to wash the nitrates to lower levels. In the seasons preceding 1928, except in 1923, most of the soluble nitrogen found in the fall was within the sampling depth of 6 feet the spring following. In 1923, however, the winter precipitation, chiefly rain, was heavy. The following spring a major part of the nitric nitrogen was found below the 6-foot level, distributed

TABLE 7

Nitric nitrogen, in pounds per acre, found to a depth of 6 feet, in plots given various treatments

TREATMENT	NITRIC NITROGEN	
	Fall	Following spring
<i>Time of plowing</i>		
Fall plowed.....	59.2	57.0
Spring plowed, volunteer 6 inches high.....	51.6	48.0
Spring plowed, volunteer 12 inches high.....	46.8	46.0
Spring plowed, volunteer in bloom.....	37.4	41.2
Spring plowed, volunteer in dough.....	34.6	37.9
<i>Manurial</i>		
Spring plowed, no manure.....	56.7	58.7
Spring plowed, 5 tons manure alternate years.....	79.0	79.1
Spring plowed, 10 tons manure alternate years.....	96.0	92.8

down to a depth of 10 feet. As a result of this movement below the rooting zone, the plants were markedly yellow, indicating a low nitrate supply, which was later reflected in subnormal yields.

In order to determine the statistical differences for the various amounts of nitric nitrogen accumulated as a result of variation in time of plowing and differences in manurial treatments, the amounts were calculated in pounds per acre. These data are shown in table 8.

For the time of plowing test, a decrease in amount of nitric nitrogen as plowing was delayed is shown with few exceptions for the individual years. The treatment means indicate the same trend, and variance analysis of the data shows that the differences were highly significant. As might be expected, the variation for years, as indicated by the ratio of the error term to the mean square for years, was also highly significant. The interaction between years

and treatments was also highly significant, indicating that the crop did not respond to the treatments similarly for all seasons.

The barnyard manure treatments included in table 8 show increased amounts of nitric nitrogen for manured plots. Statistically, it was found that the differences between treatment means as well as between yearly means were highly significant. The interaction between years and treatment was also beyond the 1 per cent level of significance.

TABLE 8

Nitric nitrogen, in pounds per acre, found to a depth of 6 feet at the beginning of the crop season after following on plots plowed at various times, and on plots treated with different amounts of barnyard manure, 1928 to 1933, inclusive

TREATMENT	NITRIC NITROGEN						
	1928	1929	1930	1931	1932	1933	Mean
<i>Time of plowing</i>							
Fall plowed.....	41.0	65.4	62.0	58.7	59.6	55.4	57.0
Spring plowed, volunteer 6 inches high.....	36.6	36.7	59.9	50.8	58.4	45.5	48.0
Spring plowed, volunteer 12 inches high.....	39.2	48.3	50.9	37.8	54.4	45.6	46.0
Spring plowed, volunteer in bloom....	32.1	34.9	41.6	39.2	44.8	54.4	41.2
Spring plowed, volunteer in dough....	28.9	32.0	50.8	37.0	33.8	44.9	37.9
Mean.....	35.4	43.4	53.0	44.7	50.2	49.1	
Differences for comparison of treatment means							
5 per cent point.....							4.7
1 per cent point.....							7.8
<i>Manurial</i>							
No manure.....	54.3	76.0	62.4	53.4	49.6	62.4	59.7
5 tons manure each alternate year....	54.2	120.4	93.4	77.8	65.4	63.6	79.1
10 tons manure each alternate year....	61.0	132.8	108.5	82.8	89.8	81.8	92.8
Mean.....	56.5	109.7	88.1	71.3	68.3	69.3	
Differences for comparison of treatment means							
5 per cent point.....							10.0
1 per cent point.....							13.8

Effect of nitric nitrogen on protein content of wheat

Protein analyses were made of the wheat harvested from the several plots involved in this study (table 9).

The results for time of plowing show that the treatment means indicate no significant trend. On the other hand, a significant difference was found to exist between yearly means. From a comparison of the yields for this test (table 3) and the amounts of nitric nitrogen (table 4) with the protein percentages (table 9), it is evident that the greater quantities of soluble nitrogen formed

as a result of earlier tillage were expressed in yield rather than in protein percentage of grain.

The barnyard manure treatments (table 9) show a definite effect upon protein percentages, as indicated by results for individual years, as well as by general treatment means. Variance analyses made of this part of the test gave proof that a highly significant difference existed between treatment means, as well as between yearly means. The interaction between years and treatments was also found to be highly significant, thus showing that the protein

TABLE 9

Percentage of protein ($N \times 5.7$) in Utah Kanred winter wheat grown on plots plowed for fallow at various times and on plots treated with different amounts of barnyard manure, 1928 to 1933, inclusive

TREATMENT	PROTEIN CONTENT OF WHEAT						
	1928	1929	1930	1931	1932	1933	Mean
<i>Time of plowing</i>							
Fall plowed.....	11.22	13.60	11.72	13.05	12.56	11.83	12.33
Spring plowed, volunteer 6 inches high.....	10.72	12.56	12.18	11.89	12.59	12.56	12.08
Spring plowed, volunteer 12 inches high.....	10.82	12.37	11.97	10.94	12.94	11.68	11.79
Spring plowed, volunteer in bloom....	11.29	13.16	12.50	11.09	13.47	12.45	12.32
Spring plowed, volunteer in dough...	10.58	13.40	12.69	11.09	13.13	11.87	12.13
Mean.....	10.93	13.02	12.21	11.61	12.94	12.08
No significant difference for treatments							
<i>Manurial</i>							
Spring plowed, no manure.....	10.73	13.12	11.92	10.34	10.58	12.31	11.5
Spring plowed, 5 tons of manure each alternate year.....	11.66	13.99	14.03	14.00	13.58	14.77	13.67
Spring plowed, 10 tons of manure each alternate year.....	13.05	14.77	14.80	16.82	14.08	17.81	15.22
Mean.....	11.81	13.96	13.58	13.72	12.75	14.96	
Differences for comparison of treatment means							
{ 5 per cent point.....							1.16
{ 1 per cent point.....							1.27

percentages were not the same each year for the various amounts of manure. The application of 5 tons of manure increased the yield from 21.3 bushels for no manure to 24.8 bushels, but the 10-ton application gave no further increase (table 3); and an average of 59.7 pounds per acre of nitric nitrogen accumulated in plots receiving no manure, 79.1 pounds in plots receiving 5 tons of manure, and 92.8 pounds in plots receiving 10 tons of manure (table 8). Apparently the extra amounts of nitrogen available in the manured plots were sufficient to stimulate yield as well as to increase significantly the protein percentage of grain.

In view of the nitric nitrogen content of each plot in these two tests at the beginning and at the end of the crop season and the nitric nitrogen percentage of the grain and straw, it is interesting to make a comparison between the amount of nitric nitrogen lost from the soil during the period of growth and the amount known to have been removed by the crop. These calculations are given in table 10.

As shown by these data, the amount of available nitrogen lost from the soil is slightly greater than that removed by the crop, which is to be expected since part of the nitrates may be utilized by soil bacteria in building up microbial protoplasm. In the manurial treatment test (table 10), it will be observed that as the amount of available nitrogen increased (table 8) the difference between the amount taken up by the crop and that lost from the soil becomes wider. In presenting this balance sheet, however, it is realized that an error is involved in the assumption that all of the nitric nitrogen lost from the soil is accounted for. Unquestionably, a certain amount which accumulated during the early growth period of the wheat was taken up by the plants. To how much this amounted obviously cannot be stated, but it was likely small as judged by the amount of soluble nitrogen accumulated on fallow plots during the same period.

Effect of nitric nitrogen on water cost of dry matter

With total crop weights, percentages of moisture at the beginning and end of the crop season, and inches of rainfall for the growth period, it is of interest to determine the water cost of dry matter for the various treatments of the time of plowing and barnyard manure tests. The water cost of dry matter (table 11) was calculated by subtracting the amount of moisture remaining to a depth of 6 feet in the soil after harvest from that found in the spring. This amount was then added to the inches of rainfall which fell from the time of sampling to the beginning of ripening. The total pounds of dry matter were then divided into the total amount of water. It is realized that in adding spring rainfall to the soil moisture, storms too limited in amount to reach the roots of the plants were included. Light showers are of value, however, in that the atmosphere is cooled, humidity is increased, and transpiration is decreased, thus making the stored soil moisture more efficient in expressing itself in crop yield.

An examination of these data shows, as might be expected, an increased water cost of dry matter as spring plowing was delayed, or as the amount of nitric nitrogen decreased as the result of delayed spring plowing. In this test, the statistical analyses showed that differences between treatment means as well as yearly means were highly significant, but only the differences between treatment means are given. In the barnyard manure test highly significant differences were also found between treatment and yearly means. As shown by the data, the water cost of dry matter decreased with the application of manure. A number of research workers have investigated the water cost

of dry matter as related to soil fertility, and in almost all cases it has been found that as the fertility increased the water cost of dry matter decreased. For manurial treatment, Montgomery and Kiesselbach (7) and Kiesselbach (5) have shown that when manure was added to soils of different degrees of fertility, the water requirement was decreased, the greatest decrease occurring with the least fertile soil. Singh and Mehta (11) found that by addition of manures the quantity of water needed for production of a unit of dry matter

TABLE 11

Water cost, in pounds, of dry matter of Utah Kanred winter wheat produced on plots plowed in fall and at various times in spring for fallow, and on plots treated with different amounts of barnyard manure for the years 1928 to 1933, inclusive

TREATMENT	WATER COST						Mean
	1928	1929	1930	1931	1932	1933	
<i>Time of plowing</i>							
Fall plowed.....	681	671	621	601	848	1,013	739
Spring plowed, volunteer 6 inches high.....	787	950	769	681	848	1,189	870
Spring plowed, volunteer 12 inches high.....	819	1,004	827	699	941	1,289	930
Spring plowed, volunteer in bloom stage.....	1,281	1,062	911	737	961	1,339	1,048
Spring plowed, volunteer in dough stage.....	1,402	1,444	1,045	831	1,131	1,432	1,214
Mean.....	994	1,026	835	710	943	1,252	
Differences for comparison of treatment means	{ 5 per cent point 116 { 1 per cent point 160						
<i>Manurial</i>							
No manure.....	482	685	665	623	847	956	709
5 tons of manure each alternate year..	466	516	533	527	776	865	614
10 tons of manure each alternate year.....	426	467	559	483	759	803	582
Mean.....	458	556	586	544	794	875	
Differences for comparison of treatment means	{ 5 per cent point 39.4 { 1 per cent point 90.9						

was reduced, but the total quantity of water transpired by the crop was increased.

SUMMARY

A study was made to determine the relationships of yield of winter wheat under dry-farm conditions to amounts of available nitrogen and of soil moisture as influenced by time of plowing and manurial treatments. From the data and discussion, the following summary and conclusions may be drawn:

Delayed spring plowing for fallow was found significantly to reduce yield of winter wheat under dry-land conditions in Utah, as the result of decreased moisture supplies and reduced amounts of nitric nitrogen.

There was no significant difference in winter wheat yields on soils treated with 5 or 10 tons of manure. The manurial treatment yields were 16 per cent higher than those on plots receiving no manure, chiefly because of the greater amounts of nitric nitrogen.

Most of the available nitrogen was used up by the time winter wheat began to ripen, but a slight increase was found later in the season, even though moisture supplies remained unchanged.

On fallow land, the accumulation of nitric nitrogen occurred in the latter part of the summer and early fall, even though soil moisture conditions were more favorable earlier in the season.

Most of the available nitrogen found at the end of the summer period in fallow land was located the following spring, but at slightly lower depth.

Time of plowing had no influence on protein percentage of winter wheat, but highly significant increases resulted from application of barnyard manure.

Loss of available nitrogen from the soil was slightly greater than that which could be accounted for by removal of the crop.

Water cost of dry matter decreased with earliness of plowing for fallow and with increased applications of barnyard manure.

REFERENCES

- (1) BRACKEN, A. F., AND CARDON, P. V. 1935 Relation of precipitation to moisture storage and crop yield. *Jour. Amer. Soc. Agron.* 27: 8-20.
- (2) BUCKMAN, H. O. 1910 Moisture and nitrate relations in dry-land agriculture. *Jour. Amer. Soc. Agron.* 2: 121-138.
- (3) CALL, L. E. 1918 The effect of different methods of preparing a seed bed for winter wheat upon yield, soil moisture, and nitrates. *Jour. Amer. Soc. Agron.* 6: 249-259.
- (4) JENSEN, C. A. 1910 Seasonal nitrification as influenced by crops and tillage. U. S. Dept. Agr. Bur. Plant Indus. Bul. 173.
- (5) KIESSELBACH, T. A. 1915 Transpiration as a factor in crop production. *Nebr. Agr. Exp. Sta. Res. Bul.* 6: 143-157.
- (6) LEATHER, J. W. 1911 Amounts of nitrogen in fallow land at Pusa, India. *India Dept. Agr. Mem., Chem. Ser.* 2: 101.
- (7) MONTGOMERY, E. G., AND KIESSELBACH, T. A. 1912 Studies in water requirements of corn. *Nebr. Agr. Exp. Sta. Bul.* 128.
- (8) SEWELL, M. C., AND GAINES, P. L. 1932 Effect of tillage treatments on soil nitrogen and carbon. *Jour. Amer. Soc. Agron.* 24: 221-227.
- (9) SEWELL, M. C., AND GAINES, P. L. 1932 Nitrate accumulation under various cultural treatments. *Jour. Amer. Soc. Agron.* 24: 283-289.
- (10) SIEVERS, F. J. 1920 Soil moisture studies. *Wash. Agr. Exp. Sta. Bul.* 155: 39-40.
- (11) SINGH, B. N., AND MEHTA, B. K. 1938 Water requirements of wheat as influenced by the fertility of the soil. *Jour. Amer. Soc. Agron.* 30: 395-398.
- (12) SMITH, J. B. 1938 Distribution of nitrates in three layers of fallow soil. *Soil Sci.* 26: 347-350.

OXIDATION-REDUCTION POTENTIALS OF ARSENATE-ARSENITE SYSTEMS IN SAND AND SOIL MEDIUMS¹

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Investigations of the effect of arsenical spray residues on orchard soils and of the effect of certain treatments on the toxicity of the arsenic residues revealed an apparent correlation between the oxidation-reduction potentials of certain treated soils and their ability to support plant growth. It was thought that a more fundamental investigation of this phase of the major problem would be of primary importance, and therefore a study was undertaken of the oxidation-reduction potentials of arsenate-arsenite systems under different soil conditions with special reference to the iron, aluminum, and colloidal clay content.

REVIEW OF LITERATURE

It has long been known that certain elements or compounds of those elements, when immersed in a solution, show a difference of potential between an electrode and the solution. This potential may be manifested either by the production of an electric current or by a chemical reaction which is characterized as an oxidation-reduction reaction upon the passage of an electric current. In this type of reaction, oxidation is considered to be the assumption of positive or the loss of negative electric charges or fundamentally the loss of electrons by atoms or ions, whereas reduction is the assumption of negative or the loss of positive charges or fundamentally the gain of electrons by atoms or ions.

In the 1890's, Peters (32) and Bancroft (3) working with this type of reaction developed a formula from the Nernst equation and the ionic theory which gives the potential of such a system when it is measured by the introduction into the system of an electrode composed of some noble metal which will not enter into the reaction. This formula, as given by Peters and subsequently known as Peters' equation, is

$$E_h = K - \frac{RT}{nF} \ln \frac{[\text{Red.}]}{[\text{Oxid.]}}$$

in which E_h is the observed potential difference between the electrode and the standard normal hydrogen electrode, and K is a constant characteristic of the particular oxidation-reduction system and is equal to the potential when the ratio of Red. to Oxid. is unity and the acid concentration is zero. This latter value is now referred to as the normal oxidation po-

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tential, designated by the symbol E^0 . The potential of the standard normal hydrogen electrode is considered to be 0. From this equation, it appears that the potential is a function only of the chemical characteristics of the system and the ratio of the concentration of the compounds capable of being oxidized or reduced. Peters assumed that the potential at the electrode is produced by the adsorption of hydrogen on the noble metal of the electrode, the hydrogen having been obtained from the reaction of the reductant on the water solvent. The specific nature and the characteristics of this type of reaction have been studied by later investigators (1, 10, 11, 12, 20, 21, 30), and the potential is now considered as arising from a perfectly reversible transfer of electrons from the oxidant to the reductant at the surface of the electrode, the latter undergoing no change and taking no part in the reaction going on in the cell. The E.M.F. under these conditions is a measure of the free energy of the cell reaction. The type of electrode that will meet the necessary requirements varies, depending on the chemical nature of the system.

In order to expedite the obtaining of a constant potential, various methods of treatment were used, such as employing the electrode in the electrolysis of dilute sulfuric acid, or placing it in a strong oxidizing agent. Other methods depend upon the use of the so-called potential mediator (16, 19, 26, 27) or the poisoning of the system by an acid or a base (12, 31).

The oxidation-reduction potential was later found to vary with the acidity of the solution, irrespective of the ratio of oxidant to reductant. This is believed to be due to the fact that the ratio of the concentrations in Peters' equation should be the ratio of the ionic concentrations instead of the molecular. Since the concentrations of the active agents are often determined by the concentration of the hydrogen ions, the latter must be taken into consideration. With the development of the Debye and Hückel activity theory, the activity of the ions rather than their concentration now is considered. Junction potentials caused by the systems used for measuring the oxidation-reduction potentials are another factor resulting in deviations of the measured values from the theoretical. These cannot be calculated and have to be either eliminated as far as possible or included in the equation constants. By the introduction of proper corrections for the acidity of the solution, the activity of the ions, and the specific nature of the system, values are obtained which correspond closely to the theoretical in those experiments in which pure solutions are used and liquid junction-potentials are at a minimum.

The exact formulas employed by different investigators vary from the simple form of Peters' equation to the complex equation.

$$E = E_{\text{obs.}} + \frac{RT}{2F} \ln \frac{c_1}{c_2} + (2T - 1) \left(\frac{RT}{F} \ln \frac{\alpha c_1}{\alpha c_2} \right) + \frac{RT}{F} \ln \frac{\alpha c_1}{\sqrt{P_{H_2}}}$$

of Noyes and Garner (29) for cerous-ceric salts. In all systems the equations developed have the same general form except for a more or less rigid definition, depending upon the type of system, the particular method employed for measuring the potential, and the object of the experiment.

Because of the complexity of the system and the difficulty of measuring and interpreting the results, only in recent years has there been a study of the oxidation-reduction potentials or of the redox potentials in soils from the viewpoint of practicality.

Neller (28) and Cooper and Wilson (14) attempted a correlation of the direct relationship between growing plants and oxidation-reduction potentials. Willis (44) and Samuel and Piper (34) suggested the possibility of the correlation of oxidation-reduction potentials of soils with iron and manganese deficiencies.

The place of oxidation-reduction potentials and reactions, in one form or another, in soil morphology has been studied by Bouyoucos (5), Sullivan and Reid (36), Gillespie (18), Shibuya, Saeki, and Ryu (35), Burrows and Cordon (9), Kononova (24), and Remesow (33).

Because of its high content of humic acids and its differential breakdown by the soil micro-organisms, soil organic matter greatly influences the oxidation-reduction potential of the soil.

Differences of opinion have been expressed, however, regarding the influence of organic matter on redox potentials. Heintze (22) claimed that organic matter causes a negative drift in the potential, whereas Burrows and Cordon (9) maintained that the drift depends on the nature of the decomposable organic matter, casein producing a strong positive potential and carbohydrates a negative value. Vandecaveye and Katznelson (38) found that the addition of Palouse litter or Helmer duff to Palouse or Helmer soil lowered the redox potential initially but that subsequently the potential increased during the period of decomposition of the organic matter. In a consideration of the potentials of nitrogenous substances, Cooper and Wilson (13) used the redox potential in an attempt to determine the different types of nitrogenous compounds likely to be present.

Brown (7) believes that the use of oxidation-reduction data will lead to a more complete understanding of both soil formation and soil processes as related to plant growth. Wartenburg (43) disagrees with previous work, because the interpretation of the oxidation-reduction potentials of soils is affected by too many factors, such as hydrogen pressure, oxygen pressure, equilibrium of reductants and oxidants of unknown character, catalysis by the metal of the electrode, and solubility, diffusibility, and nature of the reduced and oxidized phase. He claims that any correlations between soil properties and redox potentials that have been reported are due to special circumstances and to particular technics employed but are not explainable on the basis of true theoretical considerations.

Unlike the methods pertaining to work in pure solutions, those used for measuring the oxidation-reduction potentials of soils are more uniform. Of course, modifications have been adapted to fit particular conditions, but the general technics employed are similar. Bright platinum electrodes have been adopted in nearly all cases. Brown (7) measured the potential in centrifuged water suspensions of soils in which the moisture content was slightly above field conditions. Before measuring the potential, Willis (44) removed the oxygen by introducing a stream of nitrogen into the enclosed electrode chamber. Peech and Batjer (31) used a 0.1 *N* H₂SO₄ suspension in order to poise the system so that constant reproducible results would be obtained in a shorter time.

In actual interpretations of the values, the hydrogen-ion concentration, or pH, of the suspension has to be considered. Peech and Batjer (31), Herzner (23), Willis (44), Remesow (33), Vlew and Meyer (40), Heintze (22), Brown (7), and Burrows and Cordon (9) show the necessity of correcting for hydrogen ion in the soil suspension. The exact correction factor used varies. In general, the assumption is made that the electrode potential is a linear function of the pH within the limits of the system. The theoretical calculated value of 0.059 for the slope of the E_h /pH curve is given by the equation of Clark (12, p. 283)

$$E_h = E_k + \frac{RT}{nF} \ln [K_a + [H^+]] - \frac{RT}{nF} \ln \frac{[S_r]}{[S_o]}$$

in which the middle term may be reduced to

$$\frac{RT}{nF} \ln [H^+].$$

Peech and Batjer (31), however, calculated the slope of the E_h /pH curve for the soils they investigated as 0.80, and they developed several formulas showing that the slope of the curve will depend on the electrochemical equation of the oxidation-reduction reaction. Buehrer, Martin, and Parks (8) obtained their E_h /pH relationship by a dilution method in alkaline calcareous soils to which the addition of an acid or base was thought highly undesirable. The dilution of such soils permits obtaining a pH range of as much as 1.5 units by varying the extent of hydrolysis.

Volk (41, 42) has pointed out the necessity of determining the E_h /pH relationships for each soil separately and the desirability of multiple electrodes. He found that for a large

number of Alabama soils E_h determinations do not reveal whether or not an arable soil is in an oxidized or reduced state, since the E_h is dependent not only on the ratio of the oxidized to the reduced phases of the ions present, but also on the kinds and relative amounts of the ions present.

That an ionic system with a particular oxidation-reduction potential may be altered upon addition to a soil has not been denied, but the direction of its alteration has been at times devious and inexplicable.

The present work was directed to elucidating some of the factors involved in the oxidation-reduction system, arsenate-arsenite, since earlier work (39) had shown the presence of both forms of arsenic in the soil, although only the arsenate was added. The arsenate-arsenite relations are of particular interest because it has been indicated (6) that arsenite-As is more toxic to plants than is arsenate-As.

EXPERIMENTAL PROCEDURE

The colloidal fraction of the soils used in the following studies (0.002 mm. and finer) was obtained by sedimentation and centrifugation of soil water suspensions.

Quartz sand used in the experiments was purified by treatment with boiling concentrated sodium hydroxide followed by hydrochloric acid and subsequent washing with distilled water until free of chlorides. The aluminum and iron oxides used were C.P. products.

The potentials of the arsenate-arsenite system were studied in the following experiments: a series in which the ratio of arsenate to arsenite was varied by 2-mgm. increments in quartz sand, a sandy soil, and a soil of high clay content; a second series, in quartz sand, sandy soil, and clayey soil, in which the ferric oxide content was varied by 0.2-gm. increments, but the original ratio of arsenate to arsenite was established as unity; a third series, similar to the second, but in which the aluminum oxide content was varied in the same manner as was the iron oxide content; and a fourth series in sandy soil and clayey soil in which the colloidal content was increased by additions of 0.2 gm. of extracted colloid to its homologous soil with the original ratio of arsenate to arsenite as unity.

In the preparation of arsenate-arsenite systems, standard solutions of arsenic or arsenious acid dissolved in distilled water were used. In these, 1 cc. of the solution contained 1 mgm. arsenic. Soil suspensions were prepared by the addition of 20 cc. of the standard arsenic solution either as the arsenate or the arsenite to 10 gm. of soil and 20 cc. of 0.1 N H_2SO_4 . The soil suspensions were allowed to stand overnight in stoppered bottles; the following morning, they were shaken intermittently, and the potential was read, until it remained constant, by the use of a platinum electrode, a saturated calomel electrode, and a vacuum tube amplification potentiometer system.

The pH values of the suspensions were measured with a glass electrode in the same electrical system used for the redox potential determinations.

The suspensions were filtered and the arsenite, arsenate, and total arsenic in soluble forms were determined in suitable aliquots by titration with sodium bromate (2), by Zinzadze's arsenomolybdate method (45), and by the Gutzeit method (2) respectively. The silica, sesquioxides, iron, and aluminum contents of the extracted colloids were determined by the official methods of the A.O.A.C. (2).

The slope of the curve for the change in E_h with pH was determined for the two soils used. As shown in table 1, the slope closely approximates that of the theoretical value of 0.059, and therefore that correction factor was

TABLE 1
Change of the redox potential with pH in soils

SOIL	APPROX.* 0.1N H ₂ SO ₄	APPROX.* 0.1N NaOH	pH	E_h † obs.	E_h ‡
Chehalis clay	cc.	cc.			
	20	0	2.82	.544	.464
	15	5	3.69	.491	.442
	10	10	4.78	.413	.449
	0	20	6.12	.347	.463
	0	0	5.48	.384	.461
Average.....					.456
Winchester sand	20	0	1.27	.590	.418
	15	5	2.22	.532	.416
	15	15	8.24	.173	.413
	0	20	10.18	.058	.412
	0	0	6.46	.339	.474
Average.....					.427

* Ten grams of soil used.

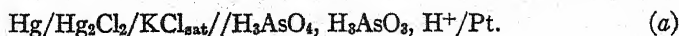
† At pH indicated.

‡ Corrected to pH of 0 and referred to normal hydrogen electrode.

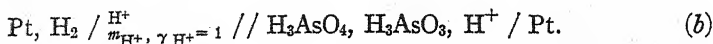
used. The column headed $E_{h \text{ obs.}}$ in this table and in the other tables gives the measured potential corrected to a pH of 2, whereas the column headed E_h gives the potential as referred to the normal hydrogen electrode, the value of 0.246 being used for the corresponding potential of the saturated calomel electrode at 21°C.

RESULTS AND DISCUSSION

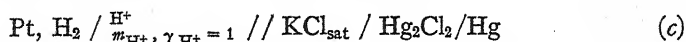
When the redox potential of an arsenate-arsenite system which is suspended in sulfuric acid is measured by the use of a platinum electrode in conjunction with a saturated calomel electrode, it may be represented (17) as being equal to the E.M.F. of the cell



In order to simplify experimental equations and calculations, the normal hydrogen electrode may be substituted for the saturated calomel electrode, in which case the cell would be



In so doing, all measured potentials would have to be corrected by the value 0.2464, which is the potential of the saturated calomel electrode as referred to the normal hydrogen electrode, this being the potential of the cell



An equation which will give the potential of the arsenate-arsenite system may be derived if 2 faradays of electricity are considered to pass through cell (b). The following reactions will take place at the electrodes:



and



with changes in chemical potential of

$$\Delta u_1 = 2u_{\text{H}^+} + 2u_{\text{e}^-} - u_{\text{H}_2} \quad (C)$$

and

$$\Delta u_2 = u_{\text{H}_3\text{AsO}_3} + u_{\text{H}_2\text{O}} - u_{\text{H}_3\text{AsO}_4} - 2u_{\text{H}^+} - 2u_{\text{e}^-} \quad (D)$$

making the total change in the chemical potential the sum of equations (C) and (D), and equal to

$$\Delta u_1 + \Delta u_2 = u_{\text{H}_3\text{AsO}_3} + u_{\text{H}_2\text{O}} + 2u_{\text{H}^+} - u_{\text{H}_3\text{AsO}_4} - u_{\text{H}_2} - 2u_{\text{H}^+} \quad (E)$$

The change in free energy of a system involving the transfer of electrons is given by Dole (15, p. 237) as

$$-\Delta G = nRT \ln \frac{P_a}{P_b} \quad (F)$$

If ΔE is the change in potential and $2F$ the number of faradays of electricity passing through cell (b), then

$$\Delta E 2F = -\Delta G \quad (G)$$

or from equation (F)

$$\Delta E = \frac{RT}{2F} \ln \frac{P_a}{P_b} \quad (H)$$

and on substituting concentrations in place of pressures in equation (H)

$$\Delta E = \frac{RT}{2F} \ln \frac{[\text{H}_3\text{AsO}_4] [\text{H}_2] [\text{H}']^2}{[\text{H}_3\text{AsO}_3] [\text{H}_2\text{O}] [\text{H}']^2} \quad (I)$$

As the voltage of a galvanic cell is regarded as the algebraic sum of the total potentials of the cell,

$$E = E_1^0 + E_2^0 + \frac{RT}{2F} \ln \frac{[\text{H}_3\text{AsO}_4] [\text{H}_2] [\text{H}^{'+}]^2}{[\text{H}_3\text{AsO}_3] [\text{H}_2\text{O}] [\text{H}^{'+}]^2} \quad (J)$$

in which E_1^0 and E_2^0 are the potentials of the standard state for each electrode. As the hydrogen electrode having the arbitrary value of 0 is used, the value E_1^0 may be eliminated. Likewise, $[\text{H}_2]$, $[\text{H}^{'+}]^2$, and $[\text{H}_2\text{O}]$ may be eliminated, since $[\text{H}_2]$ and $[\text{H}^{'+}]^2$ are constants for the normal hydrogen electrode and $[\text{H}_2\text{O}]$ remains constant for the particular concentration employed. Thus, equation (J) may be written

$$E = E^0 + \frac{RT}{2F} \ln \frac{[\text{H}_3\text{AsO}_4] [\text{H}^{'+}]^2}{[\text{H}_3\text{AsO}_3]}, \quad (K)$$

and is equal to the oxidation potential of the single arsenate-arsenite electrode. Dole (15, chap. 16), in his derivation of E^0 from the free energy concept proves that E^0 is equal to

$$\frac{RT}{nF} \ln K, \quad (L)$$

where K is the mass action constant of the electrode reaction. Thus, in the arsenate-arsenite system

$$E^0 = \frac{RT}{2F} \ln K \quad (M)$$

The redox potentials obtained by varying the ratio of arsenate to arsenite in a sulfuric acid suspension with pure quartz sand as a medium are given in table 2.

When the E_h is plotted against the logarithm of the ratio of the concentration of oxidant to reductant, as in figure 1, a straight line is obtained with a slope of 0.029, which proves that Peters' formula in its simplest form

$$E_h = E^0 + \frac{RT}{nF} \ln \frac{[\text{Oxid.}]}{[\text{Red.}]}$$

is valid for the system.

Since the total arsenic concentration in the experiment remained constant, the concentrations could not be extrapolated to zero, but by making the assumption that the ratio of the activities of arsenate to arsenite is invariable, the potential at a ratio of unity may be obtained from the curve. This potential should be equal to the normal oxidation potential of the arsenate-arsenite system, if it were not for the error introduced by the acidity of the solution.

TABLE 2

Effect of varying As_2O_5/As_2O_3 ratio on the redox potential in quartz sand, in Winchester sand, and in Chehalis clay

ORIGINAL RATIO $\frac{As_2O_5}{As_2O_3}$	$As^+ As$ As_2O_3	$As^+ As$ As_2O_3	TOTAL As	RATIO $\frac{As_2O_5}{As_2O_3}$	E_h OBS.	E_h	ΔE_h OBS.	ΔE_h CALC.
	mgm.	mgm.	mgm.					
<i>Quartz sand</i>								
0-20	0.53	19.46	0.027	0.641	0.395		
2-18	2.13	17.86	0.119	0.659	0.413		
4-16	3.86	16.13	0.239	0.665	0.419		
6-14	5.99	14.00	0.427	0.674	0.428		
8-12	8.08	11.92	0.677	0.680	0.434		
10-10	9.23	10.76	0.857	0.683	0.440		
12- 8	11.91	8.08	1.474	0.689	0.443		
14- 6	13.95	6.14	2.271	0.695	0.449		
16- 4	15.85	4.14	3.828	0.700	0.454		
18- 2	17.82	2.17	8.211	0.709	0.463		
20- 0	19.72	0.29	68.000	0.735	0.489		
<i>Winchester sand</i>								
0-20	2.26	17.68	19.94	0.128	0.572	0.326		
2-18	4.56	15.23	19.80	0.300	0.589	0.343	.017	.012
4-16	5.36	14.19	19.56	0.378	0.593	0.347	.004	.003
6-14	7.18	12.23	19.42	0.587	0.595	0.349	.002	.006
8-12	8.38	11.01	19.39	0.761	0.597	0.351	.002	.002
10-10	9.60	9.64	19.25	0.996	0.599	0.353	.002	.004
12- 8	11.92	7.18	19.11	1.661	0.602	0.356	.003	.008
14- 6	13.34	5.63	18.97	2.368	0.604	0.358	.002	.008
16- 4	14.77	4.05	18.83	3.646	0.605	0.359	.001	.003
18- 2	16.44	2.24	18.69	7.339	0.611	0.365	.006	.008
20- 0	18.12	0.43	18.55	42.139	0.620	0.374	.009	.018
<i>Chehalis clay</i>								
0-20	2.63	12.77	15.40	0.206	0.538	0.292		
2-18	3.61	10.38	14.00	0.348	0.545	0.299	0.007	0.006
4-16	4.18	9.43	13.61	0.444	0.548	0.302	0.003	0.004
6-14	4.31	8.91	13.23	0.484	0.548	0.302	0.000	0.001
8-12	4.95	7.96	12.72	0.621	0.550	0.304	0.002	0.003
10-10	5.86	6.40	12.27	0.915	0.553	0.307	0.003	0.005
12- 8	6.63	5.02	11.66	1.320	0.555	0.309	0.002	0.005
14- 6	7.33	3.88	11.21	1.889	0.557	0.311	0.002	0.004
16- 4	8.81	2.14	10.95	4.116	0.558	0.312	0.001	0.010
18- 2	9.53	1.09	10.63	8.732	0.561	0.315	0.003	0.010
20- 0	9.96	0.52	10.48	19.153	0.562	0.316	0.001	0.010

* In 40 cc. of suspension.

Thus, the value of 0.439, attained at a ratio of unity and a pH of 2, may be corrected to a pH of 0, and the value of 0.557 is obtained for the normal oxida-

tion potential of the arsenate-arsenite system at 21°C. This compares favorably with the International Critical Table value of 0.57 at a temperature of 25°C. From this potential, the value of K , the mass action constant of the

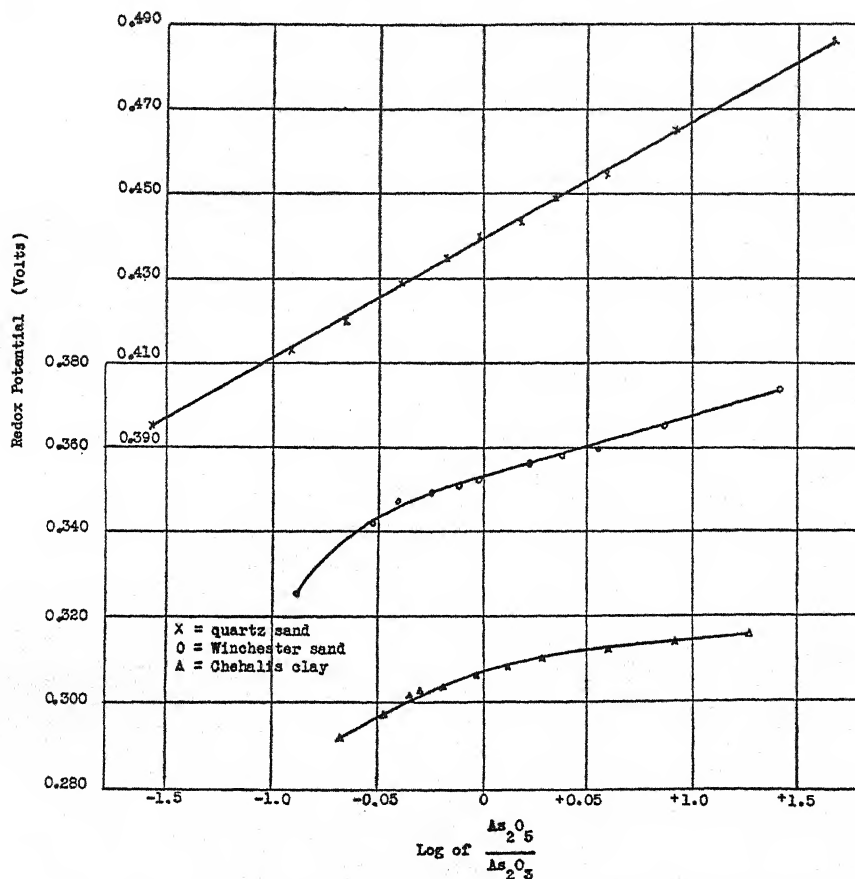


FIG. 1. INFLUENCE OF VARYING RATIOS OF ARSENATE TO ARSENITE ON THE REDOX POTENTIAL

oxidation-reduction reaction, may be calculated by substituting the value 0.56 for E^0 in formula (M)

$$E^0 = \frac{0.059}{2} \log K$$

The value thus obtained is

$$K = 9.77 \times 10^{13}$$

For the equation

$$K = \frac{[H_2AsO_4][H^+]^2}{[H_3AsO_3]}$$

This would indicate that, in the absence of other oxidation-reduction systems, at prevailing pH values in normal soils the arsenic added in the form of $\text{Pb}(\text{H}_2\text{AsO}_4)_2$ would be present predominantly as arsenate.

Because of their complexity, when soils are used as the basis for the arsenate-arsenite system, the redox potential has no significance with respect to obtaining or calculating the normal oxidation potential. A comparison of potentials, however, will reflect the oxidizing or reducing intensity of the soils.

The data for the systems in which soil is used as a medium with varying arsenate-arsenite ratios also are given in table 2. The curves in figure 1, which were obtained by plotting the measured potential against the logarithm of the ratio of the concentration of arsenate to arsenite, are parabolic and tend to form approximately straight lines with positive slopes. In the case of the clay medium, the slope of the curve is less than that of the sand medium. This may be due to the greater adsorption of arsenate arsenic by the clay than by the sand or to a lower capacity for oxidizing the arsenite to arsenate. Column 4 of table 2 discloses the fact that the fixation of arsenic per unit weight of Chehalis clay is much greater than that of Winchester sand. Of the 20 mgm. of arsenic added to the suspensions, 19.9 to 18.5 mgm. were recovered in the Winchester sand suspensions and only 15.4 to 10.5 mgm. in the Chehalis clay suspensions, depending on the ratio of arsenate to arsenite. Judged from the total quantity of arsenic recovered, a higher percentage of arsenate than arsenite is adsorbed, as indicated by the decrease in the total arsenic content when the percentage of arsenate is increased. This fixation is probably both adsorption and combination. The potentials of 0.471 for Winchester sand and of 0.425 for Chehalis clay, which are the respective values when the E_h is corrected at a ratio of unit concentration of arsenate to arsenite to a pH of 0 by means of the equation

$$E = E^0 - 0.059 \text{ pH},$$

reveal that both are reducing with respect to the arsenate-arsenite system, the clayey soil to a greater extent than the sandy soil. A comparison of the measured changes in potential caused by the variations in the arsenate-arsenite ratio with the calculated theoretical changes from the derived equation

$$E = E^0 + \frac{RT}{2F} \ln \frac{[\text{H}_3\text{AsO}_4][\text{H}^+]^2}{[\text{H}_3\text{AsO}_3]}$$

shows that the observed ΔE , in general, is greater than the theoretical at a low ratio of oxidant to reductant and smaller at a high ratio. The variations between the differences in potentials are smaller when the concentration of the arsenite arsenic is greater than that of the arsenate arsenic, as would be expected, since the apparent normal oxidation-reduction potential of both soils is lower than that of the arsenate-arsenite system.

By referring to table 1 we find that both soils are reducing with respect to the arsenate-arsenite system, but the Chehalis is less reducing than the Win-

chester. In view of the greater capacity factor of Chehalis clay, however, the equilibrium potential is lower in magnitude than that of Winchester sand. Thus, the potential of the soil system will not be directly proportional to the ratio of added oxidant to reductant. As Wartenburg (43) has pointed out, the soil system is very complex, and therefore the actual concentration and nature of the oxidant and reductant are no longer known.

The addition of an oxidizing agent such as iron to the system when the original ratio of arsenate to arsenite was unity increased the redox potential, independently of the medium used, as would be expected when one considers that the normal oxidation potential of the ferric-ferrous system is 0.74 as compared to 0.57 for the arsenate-arsenite system. Thus, there should be a tendency toward oxidation of the arsenic and reduction of the iron. The effect of the medium is reflected in the magnitude of the potential and in the existing ratios of arsenate to arsenite.

Table 3 gives the results when iron in the form of ferric oxide is added to the system. When the potential is plotted against the added concentration of Fe_2O_3 , a parabolic curve is obtained. With the initial addition of Fe_2O_3 , the potential rises sharply but soon levels off. This increase in the redox potential can be assumed to be due to an oxidation-reduction reaction between the arsenic and iron systems and to a preferential adsorption of arsenic by the Fe_2O_3 . As Biltz (4) and Lockemann and Lucius (25) have shown that arsenic in the arsenite form is much more highly adsorbed than arsenic in the arsenate form by the freshly precipitated iron hydroxide, this preferential adsorption must be taken into consideration. ✓

The pH values of the media as shown in table 3 indicated that the Fe_2O_3 employed was comparatively inert, since the acid added was not appreciably consumed. Thus the principal effect apparently resided in the solid phase Fe_2O_3 . The increase in the potential per unit addition of Fe_2O_3 was less in the soil medium than in the sand medium, as would be expected from their chemical natures and buffering capacities. The greater the colloidal content of the medium, the less was the intensity of the oxidation of arsenite upon the additions of equal weights of Fe_2O_3 . The percentage increase of arsenate arsenic caused by the oxidation of arsenite to arsenate and the preferential adsorption of arsenite arsenic was 6.63 per cent in the quartz sand, 4.34 per cent in Winchester sand, and 1.15 per cent in Chehalis clay upon the addition of 1 gm. of Fe_2O_3 . This change in the percentage oxidation is probably due to two factors: a more stabilized system, resulting from the amount and nature of the colloidal fraction, and the potential of the medium.

Aluminum oxide, as shown in table 4, had no influence on the redox potential of the system, regardless of the medium and had only a slight influence on the fixation of arsenic. The numerical values of the potential of the system and of the ratio are different in each system because of the differences in the character and nature of the medium. The fact that the potentials and the ratios remain constant regardless of the aluminum concentration proves that the

TABLE 3

Effect of varying content of Fe_2O_3 on the $\text{As}_2\text{O}_5/\text{As}_2\text{O}_3$ ratio and on the redox potential in quartz sand, in Winchester sand, and in Chehalis clay*

Fe_2O_3	$\text{As}_2^\dagger \text{ AS As}_2\text{O}_3$	$\text{As}_2^\dagger \text{ AS As}_2\text{O}_3$	TOTAL † As	RATIO $\frac{\text{As}_2\text{O}_5}{\text{As}_2\text{O}_3}$	E_h OBS.	E_h	pH
gm.	mgm.	mgm.	mgm.				
<i>Quartz sand</i>							
0.0	9.27	10.73	20.01	0.863	0.683	0.437	1.37
0.2	9.34	10.17	19.52	0.918	0.695	0.449	1.34
0.4	9.21	9.79	19.00	0.940	0.703	0.457	1.31
0.6	9.25	9.24	18.44	1.001	0.708	0.462	1.39
0.8	9.35	8.56	18.01	1.092	0.712	0.466	1.37
1.0	9.05	8.44	17.50	1.072	0.715	0.469	1.32
1.2	9.06	7.95	17.02	1.139	0.718	0.472	1.32
1.4	9.06	7.43	16.50	1.219	0.720	0.474	1.37
1.6	9.08	6.91	17.00	1.314	0.724	0.478	1.40
1.8	9.12	6.37	15.50	1.431	0.729	0.483	1.37
2.0	8.93	6.06	15.00	1.473	0.740	0.494	1.37
<i>Winchester sand</i>							
0.0	9.50	9.71	19.21	0.978	0.597	0.351	1.69
0.2	9.47	9.30	18.88	1.018	0.605	0.359	1.68
0.4	9.44	8.82	18.27	1.070	0.611	0.365	1.59
0.6	9.30	8.51	17.81	1.092	0.614	0.368	1.59
0.8	9.25	8.20	17.46	1.128	0.616	0.370	1.63
1.0	8.98	7.90	16.88	1.136	0.618	0.372	1.59
1.2	8.81	7.57	16.38	1.164	0.619	0.373	1.65
<i>Chehalis clay</i>							
0.0	5.93	6.46	12.40	0.917	0.553	0.307	2.32
0.2	5.89	6.24	12.13	0.943	0.568	0.322	2.42
0.4	5.40	5.66	11.07	0.954	0.574	0.328	2.34
0.6	5.14	5.34	10.49	0.962	0.578	0.332	2.32
0.8	4.99	5.14	10.13	0.970	0.582	0.336	2.32
1.0	4.66	4.77	9.43	0.976	0.586	0.340	2.32
1.2	4.47	4.55	9.02	0.982	0.589	0.343	2.27
1.4	4.09	4.13	8.23	0.990	0.593	0.347	2.27
1.6	4.09	4.11	8.20	0.995	0.595	0.349	2.32
1.8	3.87	3.87	7.74	1.000	0.598	0.352	2.32
2.0	3.57	3.55	7.12	1.005	0.599	0.353	2.34

* Original ratio of $\text{As}_2\text{O}_5/\text{As}_2\text{O}_3 = 1.0$.

† In 40 cc. of suspension.

slight fixation of arsenic is adsorption rather than chemical combination. If it had been the latter, there would have been a change in the ratio of oxidant to reductant. Because of the constancy of the potential and of the ratio of

TABLE 4

Effect of varying content of Al_2O_3 on the As_2O_5/As_2O_3 ratio and on the redox potential in quartz sand, in Winchester sand, and in Chehalis clay*

Al_2O_3	As† as As_2O_5	As† as As_2O_3	TOTAL† As	RATIO $\frac{As_2O_5}{As_2O_3}$	E_h OBS.	E_h
gm.	mgm.	mgm.	mgm.			
<i>Quartz sand</i>						
0.0	9.29	10.70	20.00	0.868	0.683	0.437
0.2	9.22	10.62	19.85	0.868	0.684	0.438
0.4	9.16	10.53	19.70	0.869	0.685	0.439
0.6	9.10	10.44	19.55	0.871	0.685	0.439
0.8	9.01	10.38	19.40	0.868	0.683	0.437
1.0	8.95	10.28	19.24	0.870	0.684	0.438
1.2	8.81	10.17	18.98	0.866	0.683	0.437
1.4	8.79	10.14	18.93	0.867	0.684	0.438
1.6	8.77	10.11	18.88	0.867	0.684	0.438
1.8	8.75	10.09	18.85	0.867	0.684	0.438
2.0	8.68	10.00	18.68	0.868	0.685	0.439
<i>Winchester sand</i>						
0.0	9.50	9.75	19.26	0.974	0.597	0.351
0.2	9.48	9.73	19.24	0.974	0.598	0.352
0.4	9.50	9.71	19.21	0.978	0.597	0.351
0.6	9.45	9.68	19.14	0.976	0.597	0.351
0.8	9.42	9.64	19.07	0.977	0.598	0.352
1.0	9.35	9.55	18.90	0.979	0.597	0.351
1.2	9.21	9.44	18.66	0.975	0.597	0.351
1.4	9.18	9.40	18.58	0.976	0.598	0.352
1.6	9.15	9.36	18.51	0.977	0.597	0.351
1.8	9.12	9.34	18.47	0.976	0.597	0.351
2.0	9.11	9.34	18.46	0.975	0.598	0.352
<i>Chehalis clay</i>						
0.0	6.02	6.58	12.60	0.915	0.553	0.307
0.2	5.93	6.48	12.42	0.915	0.553	0.307
0.4	5.82	6.38	12.21	0.912	0.553	0.307
0.6	5.80	6.34	12.14	0.915	0.554	0.308
0.8	5.75	6.29	12.04	0.914	0.554	0.308
1.0	5.70	6.24	11.95	0.914	0.553	0.307
1.2	5.66	6.20	11.86	0.913	0.554	0.308
1.4	5.57	6.09	11.66	0.914	0.553	0.307
1.6	5.52	6.04	11.56	0.913	0.553	0.307
1.8	5.48	5.99	11.48	0.915	0.552	0.306
2.0	5.27	5.76	11.04	0.915	0.553	0.307

* Original ratio of $As_2O_5/As_2O_3 = 1.0$.

† In 40 cc. of suspension.

oxidant to reductant, the normal potential of the arsenate-arsenite electrode may be obtained by solving for E^0 in the equation,

$$E = E^0 + 0.0295 \log \frac{\text{As}_2\text{O}_5}{\text{As}_2\text{O}_3} - 0.059 \text{ pH},$$

using concentrations instead of activities and correcting to a pH of 0. The error thus introduced is less than the experimental error. The values obtained are 0.557, 0.468, and 0.423, respectively, for quartz sand, Winchester sand, and Chehalis clay. These values agree with those obtained in the pure arsenate-arsenite systems.

The effects of additions of colloids, as may be noted from the data in table 5, are interesting from the viewpoint of behavior of the soil mediums. In both soils, the colloidal fraction possessed a greater reducing capacity than the soil from which it was extracted. With increasing colloidal contents the redox potentials in both systems decreased with a corresponding reduction of the arsenate to arsenite. The actual measured change in the potential is much greater than the theoretical change, calculated from equation

$$E = E^0 + \frac{RT}{2F} \ln \frac{[\text{H}_3\text{AsO}_4] [\text{H}^+]^2}{[\text{H}_3\text{AsO}_3]},$$

showing that the reduction in the potential is due to the increase in the colloidal content of the medium as well as to the decrease in the ratio of the concentrations of arsenate and arsenite. The reducing capacity of the two extracted colloids was similar. The average slopes of the two curves obtained by plotting the redox potential against the weight of added colloid are -0.025 for the colloid of Winchester sand and -0.026 for that of Chehalis clay. Since the redox potentials of the two soils differ initially, it appears that the potential is dependent not only on the colloidal content but also on the chemical nature of the soil material and its particle size, the chemical nature being of greater importance. The exact chemical nature of the two colloids differs also even though they have the same capacity for altering the redox potential. This is evident from the amount of arsenic fixed per gram of colloid. The colloid extracted from Chehalis clay fixed 1.99 mgm. of arsenic per gram, whereas the amount fixed by the colloid from Winchester sand was 2.6 mgm. This difference in fixation of arsenic between the two soils may be explained in part by the following facts:

The silica-sesquioxide ratios of the two extracted colloids—2.6 for Chehalis clay and 3.1 for Winchester sand—do not vary significantly, but the iron and aluminum contents of the two colloids differ markedly. The colloid from Winchester sand contains 6.02 per cent Fe_2O_3 and 22.76 per cent Al_2O_3 , whereas that from Chehalis clay contains 2.66 per cent Fe_2O_3 and 28.34 per cent Al_2O_3 . The experiments with iron and aluminum as mediums show that iron fixes more arsenic than aluminum, 2.5 mgm. of arsenic being fixed by 1 gm. of Fe_2O_3 ,

and 0.16 mgm. by 1 gm. of alumina. Not only the difference in the amount of fixation, but also the nature of the fixation must be considered. The aluminum fixation appears to be mainly adsorption, whereas the iron fixation is a combination of adsorption and chemical transformation. The variation in the iron and aluminum content of the two extracted colloids, therefore,

TABLE 5

Effect of varying content of Winchester and Chehalis soil colloids on the As_2O_5/As_2O_3 ratio and on the redox potential in homologous soils*

COLLOID	As [†] AS As ₂ O ₅	As [†] AS As ₂ O ₃	TOTAL [†] As	RATIO $\frac{As_2O_5}{As_2O_3}$	E _h OBS.	E _h	ΔE _h OBS.	ΔE _h CALC.
gm.	mgm.	mgm.	mgm.					
<i>Winchester sand</i>								
0.0	9.51	9.71	19.22	0.979	0.598	0.352	0.017	0.000
0.2	9.16	9.53	18.70	0.961	0.581	0.335	0.004	0.001
0.4	8.77	9.43	18.21	0.930	0.577	0.331	0.004	0.001
0.6	8.31	9.38	17.70	0.886	0.573	0.327	0.005	0.000
0.8	7.92	9.29	17.22	0.853	0.568	0.322	0.005	0.000
1.0	7.59	9.12	16.71	0.832	0.563	0.317	0.004	0.001
1.2	7.31	8.88	16.20	0.823	0.559	0.313	0.004	0.000
1.4	6.87	8.84	15.72	0.777	0.555	0.309	0.004	0.001
1.6	6.34	8.07	15.21	0.785	0.551	0.306	0.004	0.001
1.8	5.99	8.76	14.76	0.684	0.547	0.301	0.004	0.001
2.0	5.69	8.43	14.12	0.674	0.544	0.298	0.003	0.000
<i>Chehalis clay</i>								
0.0	5.92	6.47	12.40	0.914	0.553	0.307	0.007	0.000
0.2	5.61	6.27	11.89	0.895	0.546	0.300	0.006	0.001
0.4	4.98	5.70	10.68	0.873	0.540	0.294	0.004	0.000
0.6	4.81	5.62	10.43	0.855	0.536	0.290	0.010	0.000
0.8	4.71	5.61	10.32	0.839	0.526	0.280	0.005	0.001
1.0	4.58	5.58	10.17	0.821	0.521	0.275	0.006	0.000
1.2	4.46	5.53	10.00	0.806	0.515	0.269	0.004	0.000
1.4	4.21	5.38	9.60	0.783	0.511	0.265	0.004	0.001
1.6	3.89	5.32	9.21	0.731	0.507	0.261	0.003	0.001
1.8	3.57	5.23	8.80	0.682	0.504	0.258	0.003	0.001
2.0	3.29	5.12	8.42	0.642	0.501	0.255		

* Original ratio of $As_2O_5/As_2O_3 = 1.0$.

† In 40 cc. of suspension.

undoubtedly explains in part not only the higher fixation of arsenic but also the greater potential of the system with Winchester colloid as the medium.

CONCLUSION

The potential of an arsenate-arsenite system in a 0.05 *N* H₂SO₄-quartz medium conforms with the theoretical formula for the potential of an oxidation-reduction system. When another component is added, such as iron oxide, the

trend of the potential may be obtained from theoretical considerations of the components. On the substitution of a 0.05 *N* H₂SO₄-soil medium, there is an interference due to the complexity of the medium, and consequently the potential is no longer a direct function of the arsenate-arsenite system alone. The complexity of the soil medium, because of the many different oxidizing or reducing compounds that may be present in it and that are unmeasurable in many instances, is such that the potential can be considered only as representing the particular soil system as a whole, irrespective of any individual oxidants or reductants. The addition of special individual systems will influence the potential, and this influence may be estimated and predicted by a consideration of the potential of the soil and of the individual system. The exact numerical value and relationship, however, are often unattainable.

The addition of iron oxide to an arsenate-arsenite system results in oxidation of arsenite to arsenate with a subsequent increase in the redox potential. The potential and the percentage of oxidation vary, depending upon the nature of the soil medium. The greater the colloidal content of the soil, the lower is the potential and the smaller the percentage oxidation.

The colloidal content of the soil has a distinct bearing on the potential of the soil, but varies for each soil, depending upon the nature and percentage of the colloidal complex and the nature of the soil as a whole. The extracted colloids of the two soils studied possessed a lower potential than their homologous soils.

The oxidative character and behavior of the soil will be reflected by the redox potential, especially with reference to the addition of other chemical systems and to processes that may be going on in the soil complex. Because of the complexity of the soil medium, however, the potential cannot be considered as that of an individual system but must be regarded as a function of many complex interlocking systems. With this in mind, the redox potential may be used in the study and interpretation of the general chemical processes taking place in the soil.

The effect of certain treatments on the arsenical spray residues in soils may be interpreted on this basis. Both published (37) and unpublished results obtained in this laboratory show that additions of iron to soils poisoned by arsenical spray residues are beneficial to plant growth. Unpublished data reveal significant differences in the effects of treatments with various forms of iron. Barley grown in pots filled with arsenic-poisoned orchard soil produced 2.17, 2.43, and 4.90 gm. of dry plant material as a result of treatments with FeS, FeSO₄, and Fe₂(SO₄)₃, respectively, applied to the soil at a rate equivalent to 2000 pounds of Fe per acre. As has been pointed out in connection with the data on varying arsenate-arsenite ratios, arsenate is fixed to a higher degree by the soil than is arsenite. Thus, the observed difference in growth resulting from different forms of iron may be explained by the increased oxidation of arsenic to the arsenate form by the oxidized forms of iron. The redox potentials of the treated soils reflect the oxidation-reduction potential of the respective iron salts, that treated with FeS being the lowest and that with Fe₂(SO₄)₃

the highest. It appears, therefore, that treatments with iron in the oxidized form will result in a more intensive oxidation and a greater fixation of arsenic by the soil, with a more improved consequent plant growth, as compared with treatments with similar quantities of iron in the reduced form.

The results of the study of oxidation-reduction potentials of arsenate-arsenite systems in soil mediums suggest that the treatment of arsenic-poisoned soils with some agent, preferably one mildly oxidizing and at the same time capable of arsenic fixation, such as iron, should produce beneficial effects on plant growth.

SUMMARY

The study of arsenate-arsenite systems under different conditions disclosed the fact that Peters' formula

$$E = E^0 - \frac{RT}{nF} \ln \frac{[\text{Red.}]}{[\text{Oxid.}]}$$

is valid for the arsenate-arsenite system under ideal conditions.

Arsenite was oxidized to arsenate upon the addition of ferric oxide, and a corresponding increase in the redox potential took place.

The addition of alumina produced no effect on oxidation or reduction. The redox potential and the ratio of arsenate to arsenite remained constant.

The redox potential in soils is independent of any individual oxidant or reductant and can be considered as following the ideal equation only when the concentration of oxidant and reductant is considered to be the sum of the concentrations of all oxidizing and reducing elements or compounds in the soil, and E^0 the algebraic sum of their individual potentials. As the soil and, especially, the colloidal fraction are so complex that very few individual oxidants or reductants can be identified, the potential must be considered as representing a complex unit.

The redox potential may be used in the study and interpretation of the general nature of the chemical processes in the soil and of the changes in these processes as affected by some external factor.

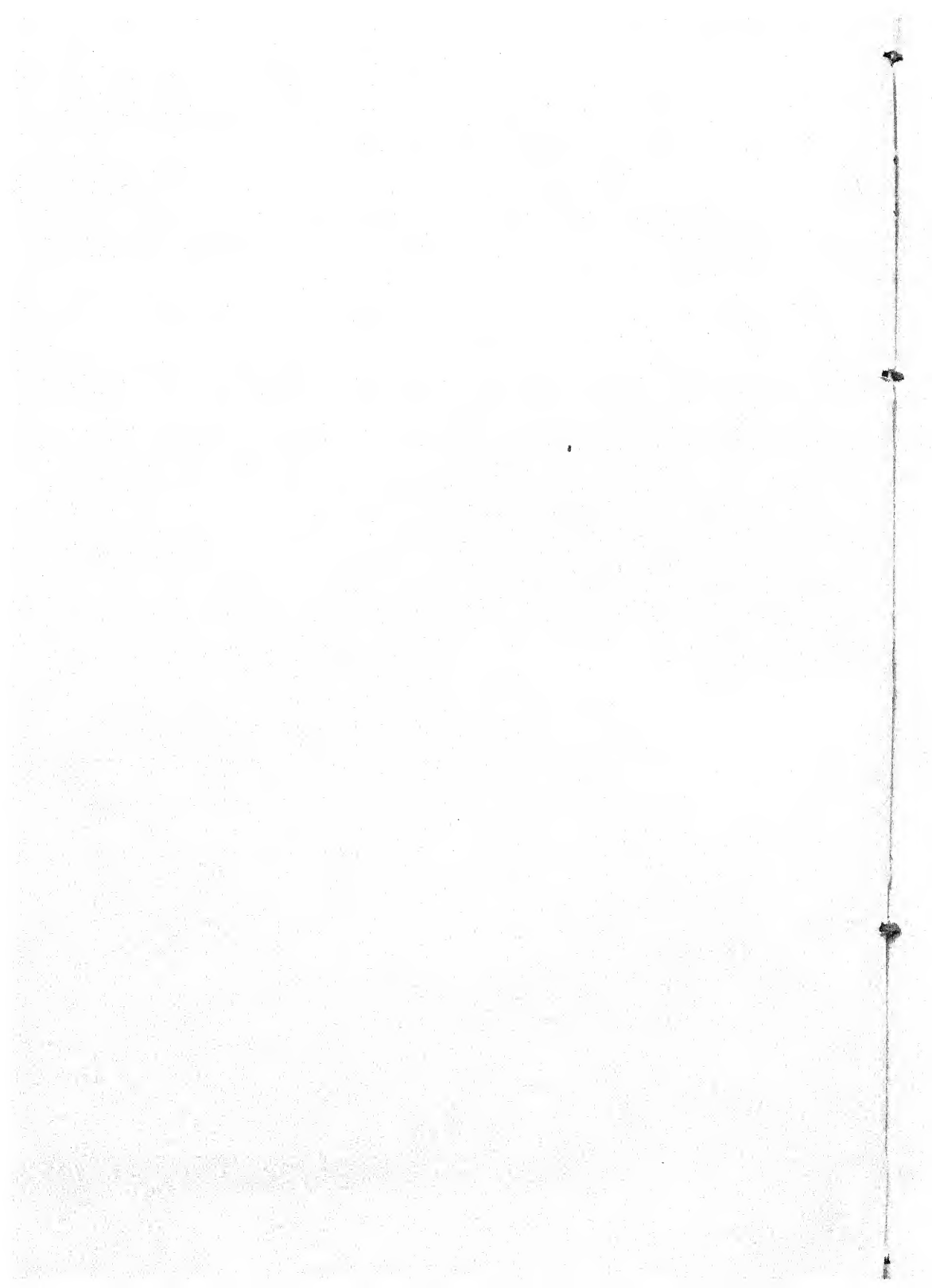
The beneficial effects of iron treatments on soils poisoned by arsenical spray residues appear to be due primarily to arsenic fixation. The total fixation was increased by oxidizing the arsenic to the arsenate form.

REFERENCES

- (1) ABEGG, R. 1904 Elektrodenvorgänge und Potentialbildung bei minimalen Ionenkonzentrationen. *Ztschr. Elektrochem.* 10: 607-609.
- (2) Association of Official Agricultural Chemists 1935 Official and Tentative Methods of Analysis, ed. 4. Washington, D. C.
- (3) BANCROFT, W. D. 1894 Über Oxydationsketten. *Ztschr. Phys. Chem.* 10: 387-409.
- (4) BILTZ, W. 1904 Über die Einwirkung Arseniger Säure auf Frisch Gefälltes Eisenhydroxid. *Ber. Deut. Chem. Gesell.* 37: 3138-3150.
- (5) BOUYOUCOS, G. J. 1923 Effect of chemical agents on oxidation in soil-forming rocks and minerals. *Soil Sci.* 15: 19-22.

- (6) BRENCHELY, W. E. 1914 Inorganic Plant Poisons and Stimulants. Cambridge University Press.
- (7) BROWN, L. A. 1934 Oxidation-reduction potentials in soils: I. Principles and electrometric determination. *Soil Sci.* 37: 65-76.
- (8) BUEHRER, T. F., MARTIN, W. P., AND PARKS, R. Q. 1939 The oxidation-reduction potential of alkaline calcareous soils in relation to puddling and organic matter decomposition. *Jour. Amer. Soc. Agron.* 31: 903-914.
- (9) BURROWS, W., AND CORDON, T. C. 1936 The influence of the decomposition of organic matter on the oxidation-reduction potential of soils. *Soil Sci.* 42: 1-10.
- (10) BUTLER, J. A. V. 1924 Studies in heterogeneous equilibria: III. A kinetic theory of reversible oxidation potentials at inert electrodes. *Trans. Faraday Soc.* 19: 734.
- (11) BUTLER, J. A. V., HUGH, W. E., AND HEY, D. H. 1926 A note of the effect of the electrode material on oxidation potentials. *Trans. Faraday Soc.* 22: 24-26.
- (12) CLARK, W. M. 1923 The Determination of Hydrogen Ions, ed. 2. Williams & Wilkins Company, Baltimore.
- (13) COOPER, H. P., AND WILSON, J. K. 1927 Correlation between electromotive series and oxidation potentials and plant and animal nutrition. *Science* 66: 629-631.
- (14) COOPER, H. P., AND WILSON, J. K. 1930 Relation of ash constituents of pasture plants to oxidation-reduction potentials of nutrients. *Soil Sci.* 30: 421-430.
- (15) DOLE, M. 1935 Principles of Experimental and Theoretical Electrochemistry. McGraw-Hill Book Company, Inc., New York.
- (16) FOERSTER, F., AND PRESSPRICH, H. 1927 Über das Electromotorische Verhalten von Arsensäure-arsenigsäurelösungen. *Ztschr. Elektrochem.* 33: 176-181.
- (17) GETMAN, F. H., AND DANIELS, F. 1931 Outlines of Theoretical Chemistry, ed. 5. John Wiley and Sons, Inc., New York.
- (18) GILLESPIE, L. J. 1920 Reduction potentials of bacterial cultures and of water-logged soils. *Soil Sci.* 9: 199-216.
- (19) GLASSSTONE, S. 1930 The Electrochemistry of Solutions. D. Van Nostrand Company, Inc., New York.
- (20) GOARD, A. A., AND RIDEAL, E. K. 1924 On the measurement of certain inaccessible potentials with a controlled hydrogen electrode. *Trans. Faraday Soc.* 19: 740-747.
- (21) HABER, F. 1904 Zeitgrößen der Komplexbildung, Komplexkonstanten, und Atomische Dimension. *Ztschr. Elektrochem.* 10: 433-436.
- (22) HEINTZE, S. G. 1934 The use of the glass electrode in soil reaction and oxidation-reduction potential measurements. *Jour. Agr. Sci.* 24: 28-41.
- (23) HERZNER, R. A. 1930 Beiträge zur Physikalischen Chemie unserer Kulturboden: III. Adsorption and Redoxpotential. *Ztschr. Pflanzenernähr., Düngung, u. Bodenk.* 18A: 249-265.
- (24) KONONOVA, M. M. 1932 Oxidation-reduction potentials as a method of characterizing conditions of soil under various methods of irrigation. *Pedology* 27: 365-376.
- (25) LOCKEMANN, G., AND LUCIUS, F. 1913 Über die Total Adsorption von Arsen durch Eisenhydroxid. *Ztschr. Phys. Chem.* 33: 735-758.
- (26) LOIMARANTA, V. L. 1907 Über die Messung Unzugänglicher Potentials durch Potentialvermittler. *Ztschr. Elektrochem.* 13: 289.
- (27) LUTHER, V. R. 1907 Zur Technik der Bestimmung von Potentialen mit unangreifbaren Electroden. *Ztschr. Elektrochem.* 13: 289.
- (28) NELLER, J. R. 1922 The influence of growing plants upon oxidation processes in the soil. *Soil Sci.* 13: 139-155.
- (29) NOYES, A. A., AND GARNER, C. S. 1936 Strong oxidizing agents in nitric acid solutions: I. Oxidation-reduction potentials of cerous-ceric salts. *Jour. Amer. Chem. Soc.* 58: 1265-1268.

- (30) PARTINGTON, J. R., AND SKEEN, J. R. 1934 Oxidation-reduction potentials in non-aqueous solutions: I. The systems cuprous-cupric chlorides and ferrous-ferric chlorides in pyridine. *Trans. Faraday Soc.* 30: 1062-1071.
- (31) PEECH, M., AND BATJER, L. P. 1935 A critical study of the methods for measuring oxidation-reduction potentials of soils, with special reference to orchard soils. N. Y. (Cornell) Agr. Exp. Sta. Bul. 625.
- (32) PETERS, R. 1898 Über Oxidations und Reduktionsketten und der Einfluss complexen Ionen auf ihre Elektromotische Kraft. *Ztschr. Phys. Chem.* 26: 193-236.
- (33) REMESOW, V. N. P. 1929 Die Oxydierenden und Reduzierenden Prozesse in dem Podzollboden. *Ztschr. Pflanzenernähr., Düngung. u. Bodenk.* 15A: 34-44.
- (34) SAMUEL, G., AND PIPER, C. S. 1929 Manganese as an essential element for plant growth. *Ann. Appl. Biol.* 16: 493-524.
- (35) SHIBUYA, K., SAEKI, H., AND RYU, K. 1936 The change in oxidation-reduction potentials of water-logged soils: II. Lateritic soils and sandstone-shale soils. *Jour. Agr. Chem. Soc. Japan.* 12: 62-73.
- (36) SULLIVAN, M. X., AND REID, F. R. 1911 Oxidation in soil. *Jour. Indus. and Engin. Chem.* 3: 25-30.
- (37) VANDECAVEYE, S. C., HORNER, G. M., AND KEATON, C. M. 1936 Unproductiveness of certain orchard soils as related to lead arsenate spray accumulations. *Soil Sci.* 42: 203-214.
- (38) VANDECAVEYE, S. C., AND KATZNELSON, H. 1938 Microbial activities in soil: V. Microbial activity and organic matter transformation in Palouse and Helmer soils. *Soil Sci.* 46: 139-167.
- (39) VANDECAVEYE, S. C., KEATON, C. M., AND KARDOS, L. T. 1938 Some factors affecting the toxicity of arsenical spray accumulations in the soil. *Proc. Ann. Meeting Wash. Hort. Assoc.* 34: 150-158.
- (40) VLES, F., AND MEYER, L. 1932 Technic of the platinum potential and apparent rH of soils. *Arch. Phys. Biol.* 9: 284-289.
- (41) VOLK, N. J. 1939 The determination of redox potentials of soils. *Jour. Amer. Soc. Agron.* 31: 344-351.
- (42) VOLK, N. J. 1939 The oxidation-reduction potentials of Alabama soils as affected by soil type, soil moisture, cultivation, and vegetation. *Jour. Amer. Soc. Agron.* 31: 577-589.
- (43) WARTENBURG, V. H. 1935 Über die Katalytischen Effekte des Platins und des Goldes in Bodenaufschlammungen und über das "Redoxpotential" des Bodens. *Ztschr. Pflanzenernähr., Düngung., u. Bodenk.* 37: 149-174.
- (44) WILLIS, L. G. 1932 Oxidation-reduction potentials and the hydrogen-ion concentration of a soil. *Jour. Agr. Res.* 45: 571-515.
- (45) ZINZADZE, C. H. 1935 Colorimetric methods for determination of arsenic in phosphorous-free solutions. *Indus. and Engin. Chem., Analyt. Ed.* 7: 230.



FACTORS INFLUENCING AVAILABILITY OF BORON IN SOIL AND ITS DISTRIBUTION IN PLANTS¹

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It is now generally accepted that boron is essential for the growth and reproduction of certain green plants. There is also conclusive evidence that some soils are deficient in this element, and that they will not produce satisfactory yields of crops until additional amounts of it are supplied. But a number of questions have arisen which must be answered before this knowledge can be intelligently applied to field practice.

In a previous paper (2) consideration was given to the adequacy of the boron supply in several soil types and to the effect of liming these soils on the availability of their boron to plants. The present paper reports the results obtained from a continuation of that study.

EXPERIMENTAL METHODS

Large samples from the A_p horizons of two soil types were collected from New Jersey farms for use in these experiments. These soil types were: Sassafras sandy loam, from the Agricultural Experiment Station at New Brunswick, and Dunellen sandy loam, from 1½ miles east of Dunellen. Each lot of soil was made to pass a ¼-inch screen and was thoroughly mixed before use.

Ten pounds of soil were placed in each of the necessary number of 1-gallon, glazed pots (without drains). A fertilizer mixture containing C.P. sodium nitrate, ammonium sulfate, and monopotassium phosphate, in proportions yielding a ratio of 5-5-7 of nitrogen, phosphoric acid, and potash, respectively, was dissolved in water and applied as required. Half of the nitrogen was supplied as the nitrate, and half in the ammonia form.

Boron was supplied as borax ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$), or boric acid (H_3BO_3), dissolved in water. Calcium was added in the form of the hydroxide or sulfate, depending upon whether it was used as a means of controlling the pH or merely for supply a corresponding amount of the element. The calcium materials were thoroughly mixed with the soil before the crops were planted.

The White-Icicle radish and the Snowball cauliflower were used as test

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plants. The radish proved to be a good indicator plant, showing marked symptoms of boron starvation when grown under conditions of low concentration of the element. In cases of mild deficiency, the root became discolored and was rough and cracked. With greater deficiency, the leaves showed abnormal development and tended to crack at the midrib. Cauliflower was also a good indicator plant and responded quickly to applications of boron. It had the added advantage of providing large quantities of fresh tissue from which the juice could readily be pressed.

In obtaining the juice of cauliflower plants for boron determinations, 250-gm. samples of tissue were taken from the stems, heads, and leaves, respectively, and immediately frozen at temperatures between 20° and 25°C. The frozen samples were thawed, and their juice was squeezed out by means of a Carver press, using pressures between 7,000 and 8,000 pounds per square inch, for 5 minutes. The juice was then stored in glass jars at temperatures around 6°C., being filtered through a Büchner funnel before analysis for boron. All boron determinations were made by the quinalizarin method of Smith, with the modification mentioned in the previous paper.

Calcium was determined by the A. O. A. C. method. Soil pH values were measured with a Leeds and Northrup potentiometer, with a glass electrode, in a 1:1 suspension of soil and water, which had been allowed to stand for 2 hours before readings were taken.

INFLUENCE OF CALCIUM ON AVAILABILITY OF BORON

Previous work had shown that liming a soil has a marked effect in lowering the availability of its boron but did not provide a clue as to whether this was due merely to the change in pH or to the increased supply of calcium. Accordingly, equal amounts of calcium, in the form of the sulfate and the hydroxide, respectively, were added to separate series of pots. Each treatment was replicated three times. Distilled water was added in amounts equal to 30 per cent of the soil's moisture-holding capacity. The moisture supply was maintained at 30 per cent saturation for 2 weeks, after which a small sample of soil was removed for pH determinations, and radish seeds were then planted. After the radishes were harvested, another application of fertilizer was made to the soil, and a second crop of radishes was grown.

Table 1 shows the effect of hydrated lime, when added to Sassafras sandy loam, on the yield and boron content of the two successive crops of radishes. Marked differences in the growth and appearance of the roots were observed at the several pH values. Typical boron deficiencies occurred at pH values of 7.6 and higher. There was a substantial reduction in the weights of roots, at these higher pH levels, but the weight of the tops increased instead. The boron content of the crop decreased with increasing rates of applications of hydrated lime. Boron deficiency symptoms were not nearly so marked in the second crop as in the first.

The effect of increasing quantities of calcium sulfate on the yield and boron content of radishes is shown in table 2. The calcium sulfate caused a reduction

TABLE 1

Yield and boron content of radishes on Sassafras sandy loam as influenced by applications of hydrated lime

HYDRATED LIME ADDED	SOIL REACTION AT START	ROOTS		TOPS			
		Fresh	Dry	Fresh	Dry	Boron	Boron
lbs.*	pH	gm.	gm.	gm.	gm.	p.p.m.	mgm.
<i>First Crop</i>							
None	5.8	153	5.7	38	3.5	25	0.09
500	6.3	153	5.6	41	3.6	23	0.08
700	6.7	147	4.9	41	3.4	24	0.08
1,000	7.0	141	4.8	43	3.6	22	0.08
1,400	7.2	146	4.9	45	3.5	17	0.06
2,000	7.6	130	4.8	44	3.7	13	0.05
2,700	7.7	111	4.7	47	4.2	9	0.04
3,600	7.9	100	3.9	51	4.2	10	0.04
<i>Second Crop</i>							
None	6.3	85	3.0	18	1.3	38	0.05
500	6.5	87	3.3	20	1.4	40	0.05
700	6.5	91	3.3	19	1.5	30	0.04
1,000	6.6	89	3.1	20	1.5	29	0.04
1,400	6.9	86	3.3	20	1.5	27	0.04
2,000	7.0	76	3.2	18	1.7	19	0.03
2,700	7.3	74	3.0	19	1.7	21	0.03
3,600	7.6	78	3.0	23	1.8	20	0.03

* Per 2,000,000 pounds of soil.

TABLE 2

Yield and boron content of radishes on Sassafras sandy loam as influenced by applications of calcium sulfate

CALCIUM SULFATE ADDED	SOIL REACTION AT START	ROOTS		TOPS				
		Fresh	Dry	Fresh	Dry	Calcium	Boron	Boron
lbs.*	pH	gm.	gm.	gm.	gm.	per cent	p.p.m.	mgm.
<i>First Crop</i>								
0	5.8	153	5.7	38	3.5	6.5	21	0.08
1,250	5.4	124	4.9	51	4.2	6.6	47	0.19
2,500	5.4	130	5.0	51	4.3	6.9	46	0.19
5,000	5.4	124	4.9	46	4.0	7.2	35	0.14
<i>Second Crop</i>								
0	6.3	85	3.0	18	1.3	...	38	0.05
1,250	6.0	81	3.3	19	1.7	...	57	0.09
2,500	5.5	83	3.7	20	1.7	...	40	0.06
5,000	5.4	77	3.0	18	1.6	...	40	0.06

* Per 2,000,000 pounds of soil.

in the yield of radishes, but it had no influence on the supply of boron which was available to the plants. In fact, the boron content of plants growing on soils receiving moderate applications of calcium sulfate was greater than that of the plants on the untreated soil. The second crop of radishes, on the soils which were treated with calcium sulfate, did not contain as large amounts of boron as the first crop.

EFFECTS OF FOUR HYDROXIDES OF AVAILABILITY OF BORON

Consideration was given to the comparative effects of the hydroxides of calcium, magnesium, sodium, and potassium on the availability to plants of

TABLE 3

Yield and boron content of radishes on Sassafra sandy loam as influenced by hydroxides employed to control soil pH

HYDROXIDE ADDED	SOIL REACTION		BORIC ACID ADDED	ROOTS		TOPS			
	Start	End		Fresh	Dry	Fresh	Dry	Boron	Boron
	pH	pH	lbs.*	gm.	gm.	gm.	gm.	p.p.m.	mgm.
None.....	5.9	5.8	0	53	3.1	22	2.3	34	0.07
			1.5	56	3.1	23	2.2
Sodium.....	7.8	7.6	0	46	2.4	34	3.0	15	0.04
			1.5	49	2.4	36	3.1
Potassium.....	7.7	7.2	0	58	3.3	27	2.6	21	0.05
			1.5	58	3.2	28	2.7
Calcium.....	7.7	7.6	0	55	3.3	27	2.5	12	0.03
			1.5	64	3.6	27	2.7
Magnesium.....	7.7	7.2	0	41	2.5	26	2.7	9	0.02
			1.5	62	3.7	24	2.5

* Per 2,000,000 pounds of soil.

the boron in soils. Sufficient amounts of saturated solutions of the first two hydroxides, and 0.2 *N* solutions of the last two, were added to pots of soil, in groups of six, to raise the pH to approximately 7.8. Fertilizer was mixed with the soil, and radish seeds were planted.

As soon as the seedlings came up, differences in the growth and appearance of the leaves were readily apparent. Boron deficiency symptoms were observed only on the plants growing on soils which received the calcium and magnesium hydroxides. The symptoms were much more marked in the latter case than in the former.² Table 3 gives the data on the yield and boron content of the roots and tops of radishes grown in this experiment.

² It is interesting to note that Talibi (1) found high-magnesian liming materials caused a greater reduction in plant growth than did high-calcium materials, under conditions of an inadequate supply of boron.

It is evident from these data that the nature of the cation employed to regulate the pH of the soil has an important bearing on the availability of the boron in the soil. This has been borne out in solution culture tests (3) in which it was shown that a change in pH, without a corresponding change in the content of calcium or magnesium in the solution, had little effect on the absorption of boron by plants.

TABLE 4

Boron content of juices and residues of various parts of cauliflower plant as influenced by borax applications to the soil

BORAX ADDED*	PLANT MATERIAL		JUICE	RESIDUE	BORON CONTENT				
	Fresh	Dry	Total	Dry	Un-pressed	Juice	Residue	Juice	Residue
	lbs.	gm.	gm.	gm.	gm.	p.p.m.	p.p.m.	p.p.m.	mgm.
<i>Stems</i>									
0	280	28	151	18	18	0.2	26	0.03	0.46
5	333	40	162	26	21	0.3	31	0.05	0.80
10	340	45	153	29	23	0.2	33	0.03	0.95
20	382	45	168	28	24	0.3	34	0.05	0.95
40	336	35	174	23	25	0.2	36	0.03	0.82
<i>Leaves</i>									
0	370	35	155	20	14	0.2	20	0.03	0.40
5	659	80	276	54	22	0.2	28	0.05	1.51
10	915	105	356	69	26	0.5	33	0.18	1.79
20	1,065	118	426	77	31	0.6	42	0.25	3.23
40	542	45	218	29	45	1.0	55	0.21	1.59
<i>Heads</i>									
0	396	30	221	15	10	0.2	15	0.04	0.22
5	503	43	302	24	14	0.2	23	0.06	0.55
10	670	60	375	29	16	0.3	26	0.11	0.75
20	679	65	427	34	18	0.3	30	0.13	1.02
40	986	81	591	37	19	0.3	32	0.17	1.18

* Per acre of soil.

The exact mechanism of the change in availability of soil boron effected by liming is unknown. The possibility of chemical fixation of boron by lime has been pointed out by a number of investigators. Other workers have given a biological explanation for this phenomenon. The present study indicates that the former affords a plausible explanation. The boron content of the tops of the plants growing on soils to which the several hydroxides were applied varied according to the known solubilities of the corresponding metaborates.

DISTRIBUTION OF BORON IN CAULIFLOWER PLANTS

Cauliflower plants, which had been started in a rich loam soil, were transplanted into Dunellen sandy loam of low fertility. Fertilizers, equivalent to

2,000 pounds of 5-5-7, were thoroughly mixed with the soil at planting time. Additional nitrate of soda, at rates equivalent to 500 pounds per acre, was applied at intervals of 6 and 10 weeks after transplanting. Boron was supplied in the form of borax. When the heads had formed, the plants were harvested,

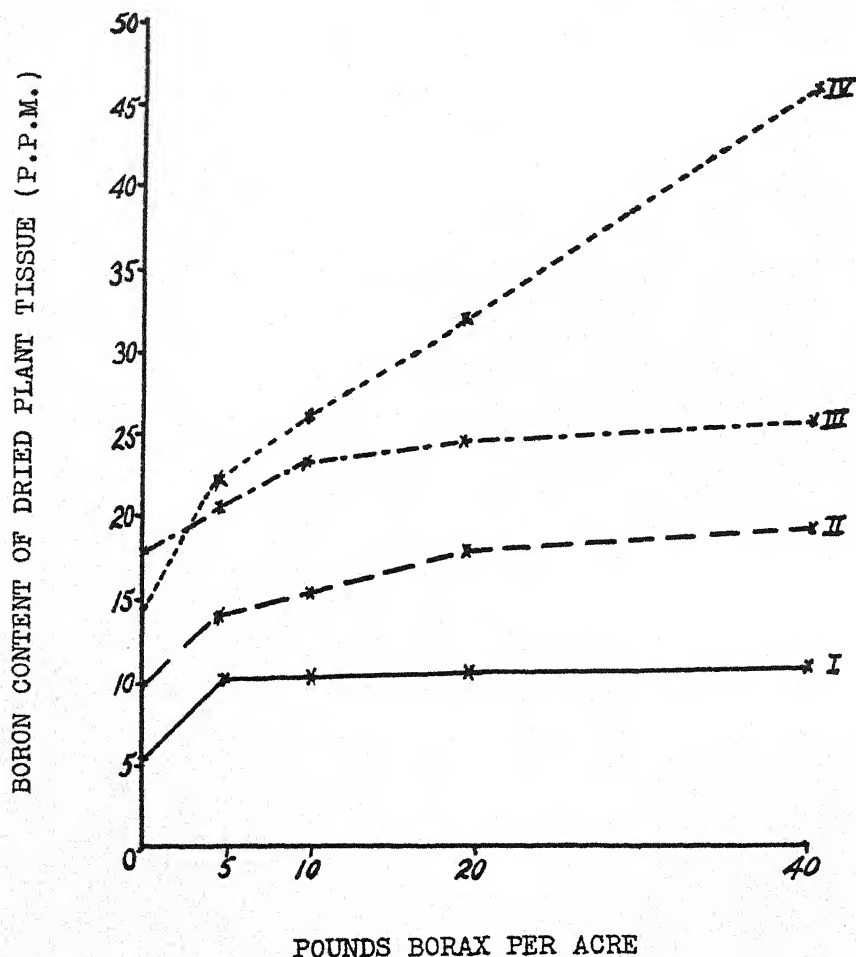


FIG. 1. BORON CONTENTS OF PARTS OF CAULIFLOWER PLANTS AS INFLUENCED BY APPLICATIONS OF BORAX TO SOIL

I = Roots; II = Heads; III = Stems; IV = Leaves

and determinations were made of the boron content of the juice, residue, and unpressed tissues of samples taken from the stems, leaves, and heads. The data are shown in table 4.

Symptoms of boron deficiency and excess were observed (plate 1) at the low and high rates of application, respectively. In plants growing on soils receiv-

ing no borax, the largest percentage of boron was found in the stems, and the least in the roots. In plants growing on soils which had been liberally treated with borax, the boron content (fig. 1) was greatest in the leaves and least in the roots. The amount of boron in the expressed juices ranged between 3 and 13 per cent of the total amounts present in the tissues. The boron concentration of the juices of the best cauliflower plants was between 0.2 and 0.5 p.p.m.

Determinations were made of the solubility in water, ethyl alcohol, and 2 per cent hydrochloric acid solution of the boron in the residue of pressed cauliflower plants. Three-gram samples of the residue were placed in boron-free glass flasks, to which 100 cc. of liquid had been added. The contents of the flasks were shaken intermittently for 1 hour and were then transferred to a paper filter and washed with 300 cc. of the extracting liquid, followed by the

TABLE 5
Boron in cauliflower plant tissue after extraction with solvents

TYPE OF TISSUE	BORAX APPLIED	TOTAL BORON*	BORON CONTENT* OF TISSUE AFTER BEING TREATED WITH:			
			Cold Water	Hot Water	Ethyl Alcohol	2 per cent Hydrochloric Acid
	lbs.†	mgm.	mgm.	mgm.	mgm.	
Roots.....	0	0.006	0.002	Trace
	40	0.010	0.004	Trace
Stems.....	0	0.018	0.006	Trace
	40	0.024	0.008	Trace
Leaves.....	0	0.012	0.011	0.003	0.010	Trace
	40	0.046	0.044	0.012	0.040	Trace
Heads.....	0	0.010	0.006	0.010	Trace
	40	0.019	0.007	0.020	Trace

* Per gram of tissue.

† Per acre of soil.

same amount of distilled water. The filter paper and its contents were placed in a crucible and ignited, and boron was determined in the ash. The data are shown in table 5.

Only traces of boron were extracted by cold water and alcohol. Considerably more of it was soluble in hot water, and practically all of it dissolved in 2 per cent hydrochloric acid solution. The solvent effects were almost identical on the several parts of the plants. This indicates that the boron stored in the tissues of plants is not available for use in the newly developing parts and explains why there must be a constant source of supply of boron in the nutrient solution. As boron enters the plant, the roots remove what they need for their development. The remainder moves up into the stems and leaves of the plant where it is rapidly immobilized.

SUMMARY

In a study of the effects of lime and fertilizers on the adequacy of the supply of boron in Sassafras sandy loam, it was found that, for radishes and cauliflower:

Boron deficiency symptoms occurred at pH levels approaching 7, and became increasingly severe at higher values.

The percentage and total amount of boron in plant tops decreased as the pH of the soil increased.

Of the four hydroxides tested, that of magnesium caused the greatest reduction in the availability of soil boron. Calcium, sodium, and potassium hydroxides had lesser effects, in the order named.

In a study of the distribution of boron in cauliflower plants grown on Dunellen sandy loam, it was found that:

Applications of 5 or 10 pounds of borax per acre overcame boron deficiency, but 20-pound applications resulted in toxicity.

Boron consumption by plants increased when additional quantities of borax were added to the soil.

The expressed juices of plants contained much less boron than the remaining plant residue.

The boron in plant residues from which the juice has been pressed is insoluble in cold water and in alcohol, but is appreciably soluble in hot water and completely soluble in 2 per cent hydrochloric acid solution.

REFERENCES

- (1) TALBI, A. 1935 Bedeutung von Mikroelementen und des Verhältnisses von Ca/Mg für das Pflanzenwachstum bei Kalkingen saurer Boden. *Ztschr. Pflanzenernährung, Düngung, u. Bodenk.* 39 A: 257-264.
- (2) WOLF, B. 1939 Experiments with boron on some New Jersey soils. *Soil Sci.* 48: 41-57.
- (3) WOLF, B. 1940 Boron: A study of its availability and adequacy in a Sassafras sandy loam soil and of its distribution in cauliflower plants. Thesis: Rutgers University, New Brunswick, N. J.

PLATE 1

CAULIFLOWER GROWN ON DUNELLEN SANDY LOAM WHICH HAD BEEN TREATED WITH BORAX
AT ACRE RATES INDICATED

FIG. 1. None.

FIG. 2. Five pounds.

FIG. 3. Ten pounds.

FIG. 4. Twenty pounds.

FIG. 5. Forty pounds.



FIG. 1

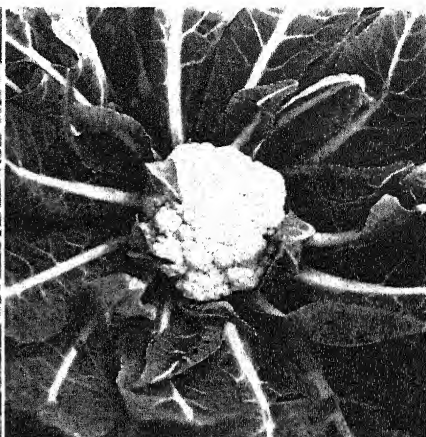


FIG. 2



FIG. 3

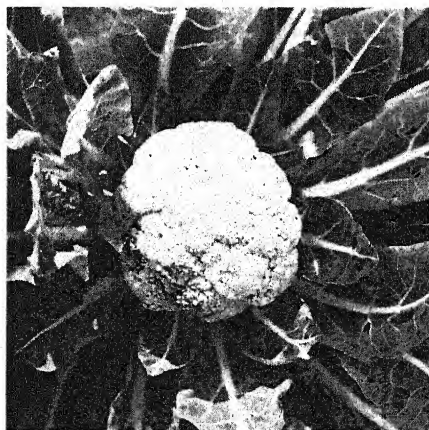


FIG. 4



FIG. 5

NATURE AND LIMING VALUE OF QUENCHED CALCIUM SILICATE SLAG¹

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Of the several calcic slags from industrial operations, "basic" (Thomas) slag has been most important to agriculture. Basic slag comes from furnace operations in which iron ore is reduced and its phosphate and silica content fluxed by admixed limestone. Although a strongly basic material and of liming value, the ground unquenched slag has been classified as a phosphatic fertilizer and is evaluated upon the basis of "available" P_2O_5 content.

Substantial quantities of a relatively new type of quenched slag now come from rock phosphate reduction furnaces. This by-product, chiefly calcium silicate, is quite different from "basic slag" in chemical composition and in physical characteristics. The present paper presents laboratory and pot culture studies of the liming value of this relatively new by-product.

LIMING EFFECTIVENESS OF CALCIUM SILICATE

Silicate minerals, wollastonite and serpentine, were compared with both mineral and precipitated carbonates of calcium and magnesium in laboratory and pot studies at the Tennessee Station in 1914 (8, 12). The two alkaline-earth silicates proved equal or superior to their analogous carbonates, upon the basis of comparisons between chemically equivalent incorporations. No record of advocacy of calcium silicate as a liming material had been found when those results were published. In September 1934, however, the late S. D. Conner supplied the following excerpt from Johnson, 1864 edition (7): "There is one compound of lime, which occurring occasionally in all soils, has not hitherto been applied to the improvement of land, even in localities where it abounds. This compound is the silicate of lime." Johnston then pointed out that in smelting iron, "the first slag that is obtained consists in great part of silicate of lime" and that "on 'boggy' and peaty soils it may be laid in large quantity," and "it will decompose slowly and while it imparts to the soil solidity and firmness, will supply both lime and silica to the growing crops, for a long period of time."

Subsequent pot culture studies have demonstrated the effectiveness of

¹ A cooperative study, The University of Tennessee Agricultural Experiment Station and Departments of Chemical Engineering and Agricultural Relations of Tennessee Valley Authority.

various calcium silicates as liming materials (1, 2, 3, 4, 5, 6, 13, 14, 15, 17, 18, 19), and theoretical considerations have been discussed by Thomas (16). An industrial by-product, "dicalcium silicate," has been compared with burnt lime and limestone in collaborative lysimeter studies at the Tennessee and Virginia Stations during the last 15 years (9). Quenched and unquenched slags from rock phosphate reduction furnaces have been subjected to laboratory and pot studies at the Tennessee Station during the last 5 years, and the behavior of their admixtures with superphosphates has been reported (10).

QUENCHED CALCIUM SILICATE SLAG

Physical nature and chemical composition

Molten slag is drawn from the bottom of the rock phosphate reduction furnace and chilled and shattered by impingement of a stream of water. The resultant

TABLE 1

Mechanical and chemical percentage analyses and neutralizing values of quenched calcium silicate slags produced at different times and places

SLAG NUMBER	MECHANICAL ANALYSIS						CHEMICAL ANALYSES*				NEUTRALIZING VALUE CaCO_3 *	
	Fractions retained by mesh designates						CaO	SiO_2	P_2O_5	F	Com-puted†	Ti-trated
	4	10	16	20	60	<60						
S-349‡	0	13.6	30.2	20.8	31.5	3.9	54.33	35.8	1.38	3.17	87.5	84.0
S-653‡	0	8.0	25.9	23.3	40.3	2.5	45.10	42.3	1.40	2.20	71.7	70.0
S-657§	0	30.0	24.6	18.0	23.6	3.8	42.61	41.7	1.25	2.47	66.9	68.0
S-686‡	0	36.0	29.2	15.1	17.9	1.8	49.84	40.0	1.84	2.37	78.9	79.0
S-693‡	0	7.3	17.1	18.5	48.4	8.7	52.08	39.9	1.30	2.30	84.2	80.0
S-792	0	18.9	33.2	19.5	24.0	4.4	50.80	40.9	1.75	2.16	81.8	79.5
S-795‡	0	1.3	10.5	14.5	57.3	16.4	52.70	34.9	1.25	3.20	84.0	79.0

* Material minus 100-mesh.

† Equivalence of total Ca minus that accounted for by P_2O_5 and F_2 .

‡ Wilson Dam product.

§ Commercial Tennessee product.

|| Commercial Florida product.

glassy product then is propelled through pipe lines to storage piles. The unground quenched material varies from white to dull gray. Particles are brittle, pitted, and sharp-edged. Mechanical analyses of table 1 show that most of the material is within the range of 10 to 60 mesh.

The physical structure of stream-quenched slag differs somewhat from that of the special material, obtained by delivery of molten slag into a body of water, used in previous studies of admixtures with superphosphates (16). Microphotographs of quenched and unquenched slags from Wilson Dam and from commercial operations in Tennessee and Florida are shown in plate 1.

The slag contains virtually all of the component elements of raw rock phosphate, except the almost completely volatilized phosphorus. Practically

all of the calcium attributable to the phosphate and carbonate in the raw rock is fluxed into chemical combination with added silica. Slags from different furnace operations vary somewhat in composition and neutralizing value, because of the variable composition of the raw rock and the consequential variation in the ratios of quartz and coke incorporated with the furnace charge. Representative "complete" analyses of slags have been given elsewhere (10), and partial analyses of seven slags from operations in Tennessee, Alabama, and Florida are given in table 1.

TABLE 2

Distilled water and carbonated water dissolutions of quenched calcium silicate slags produced at different times and places

SLAG NUMBER*	CO ₂ -FREE AQUEOUS EXTRACTS†				CARBONATED WATER EXTRACTS‡					
	pH‡	Solute alkalinity, CaCO ₃ ≡ per charge	Fluorine		CaCO ₃ ≡		P ₂ O ₅		Fluorine	
			Actual	Fraction of total in slag	Per L.	Fraction of total	Per L.	Fraction of total	Per L.	Fraction of total
		per cent	per cent	per cent	mgm.	per cent	mgm.	per cent	mgm.	per cent
S-348§	10.2	0.60	0.013	0.41
S-349§	10.8	1.30	0.032	1.01	122.5	24.12	1.8	26.0	5.01	31.6
S-653§	9.9	0.45	0.013	0.51	65.0	16.14	0.8	11.4	2.00	20.0
S-657	10.3	0.60	0.019	0.77	62.5	16.42	0.6	8.2	2.68	21.7
S-686§	9.8	0.45	0.020	0.85	60.0	13.48	1.4	15.2	1.67	14.1
S-693§	10.4	0.85	0.025	1.08	82.5	17.74	0.8	12.3	3.17	27.5
S-791**	10.8	0.67	0.025	1.20
S-792**	11.2	1.02	0.044	2.05	100.0	22.40	1.6	18.0	5.51	51.0
S-795§	10.6	1.05	0.019	0.59	60.0	12.74	0.6	9.6	1.75	10.1

Slags S-348 and S-791 were ground unquenched materials, otherwise identical, respectively, with quenched slags S-349 and S-792.

* All charges were 100-120-mesh material for both extractions.

† 1-gm. charge per 100 cc., 1 hour agitation; pH values by glass electrode; fluorine by Willard and Winter procedure (20).

‡ Four-hour agitated extraction with distilled water saturated with CO₂ at 8° C.

§ Wilson Dam product.

|| Commercial Tennessee product.

** Commercial Florida product.

The liming value of slag is expressed as the CaCO₃-equivalence of its calcium silicate content. In general, the magnesia and the monovalent bases present may be disregarded. The potential neutralizing value of the calcic components of slag can be computed as the difference between total calcium content and the amount accounted for jointly by phosphate and fluoride combinations. For ordinary requirements, however, proximate neutralizing values of slags of low phosphate content can be determined by dissolution of slag in acid and back titration. Neutralizing values of seven slags, determined by the aforementioned computation and by direct dissolution-titration technic, are given in juxtaposition in table 1.

Solubility in water and in carbonated water

Slags from different furnace operations and from the same operation vary also in their solubility in aqueous systems. The pH, titrated alkalinity, and fluoride content of CO_2 -free aqueous extracts of 100–120-mesh separates of nine slags are given in table 2. The results show that variation in $\text{CaO}:\text{SiO}_2$

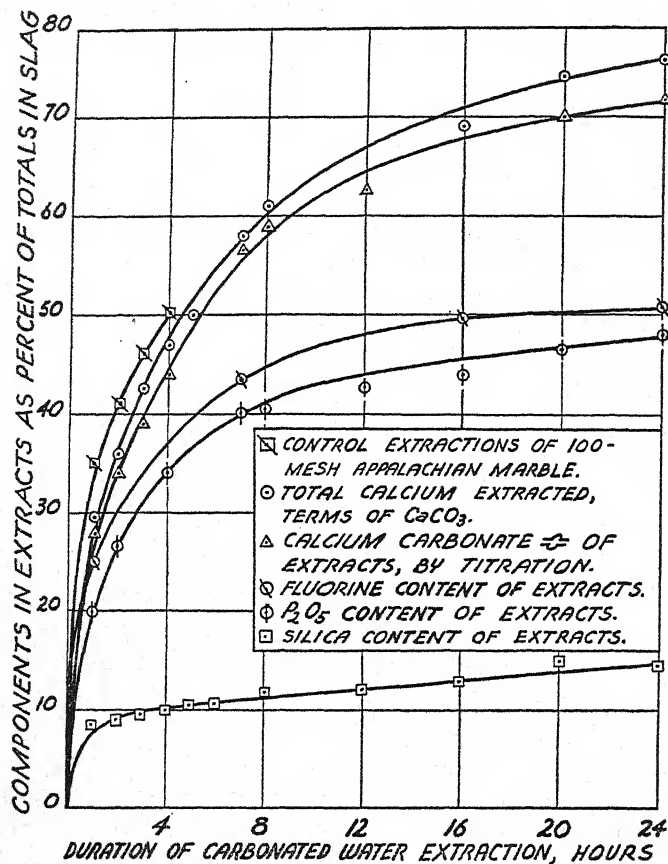


FIG. 1. PROGRESSIVE DISSOLUTION OF QUENCHED CALCIUM SILICATE SLAG IN CARBONATED WATER, AS REGISTERED BY CALCIUM, P_2O_5 , FLUORINE, AND SILICA CONTENTS OF PERIODIC EXTRACTS

ratio influences hydrolysis and dissolution of slag and that quenching increases the rate of solubility.

The efficacy of slag as a liming material is governed by the speed of its dissolution and the diffusion of the resultant solutes from slag particles in the soil. The speed of dissolution of 100-mesh Wilson Dam slag, S-349 of tables 1 and 2, and the solute content of progressive carbonated-water extracts are shown in figure 1. A slag charge of 1-gm. was introduced into a liter of car-

bonated water, saturated with CO_2 at $5^\circ\text{C}.$, and the container was sealed. Twelve such systems were agitated simultaneously in an overthrust shaking machine for periods up to 24 hours. One system was removed at the end of each of the eleven designated intervals and analyzed for total solute calcium, calcium bicarbonate, P_2O_5 , fluorine, and solvated silica. The calcium carbonate content of the undissolved fraction of each system was found to be 1 per cent, or less, of the slag charge.

The curves of figure 1 demonstrate that the calcium silicate of the slag readily undergoes hydrolysis in CO_2 -saturated aqueous systems and yields a calcium bicarbonate solution of concentration approximating that obtained by parallel dissolution of Appalachian marble of like charge and fineness. The curve for total solute calcium lies above the curve for generated calcium bicarbonate, because of concomitant dissolution of calcium phosphate and fluoride, as indicated by extractions of approximately one-half of the fluorine and P_2O_5 content of the slag. More than one-half of the quantity of each of the four components dissolved during 24 hours was accounted for by extraction during the first 3 hours.

Dissolution in CO_2 -saturated distilled water during a 4-hour period of agitation was used as a means of comparison of the seven quenched slags of table 1. Fractions of slag converted into calcium bicarbonate and percentage dissolutions of P_2O_5 and fluorine content are given in table 2.

Influence of particle size upon speed of dissolution

Quenching facilitates and expedites handling of the molten slag and obviates grinding of the rocklike air-cooled material. A corollary benefit of quenching is the production of a brittle slag of pitted surface and somewhat greater solubility. It would be advantageous economically if quenched slag could be used at ordinary rates without further mechanical breakdown. Agricultural limestone of similar particle size is too coarse, however, for effective liming at economic rates, and the less rapidly dissolved coarse quenched slag would be expected to show similar inadequacy.

Influence of particle size upon speed of dissolution in carbonated water suspensions was determined, therefore, by means of five separates of slag S-349 of table 1, in 10- to 200-mesh range. A 0.5-gm. charge of each separate was suspended in a liter of carbonated water and the suspension agitated 16 hours. Identical simultaneous treatment was accorded five corresponding separates of Appalachian marble. The curves of figure 2 show dissolved CaCO_3 values for the two series of extractions.

Dissolution of the finest separate of slag was seven times that of the coarsest separate. The percentage extraction from marble exceeded that from the slag in each comparison. The spread between the curves of each pair would be greater were solute CaCO_3 values graphed as concentrations, instead of as percentage extractions from the marble, of 99 per cent of the CaCO_3 content, and from the slag, which had a potential calcium carbonate value of 87.5 per cent.

Dissolution of pulverized wollastonite of 95 per cent purity was brought into the comparisons. The curve for dissolution of 100-mesh wollastonite was much steeper than the curve for 100-120-mesh marble and also topped the corresponding curve for the 200-mesh separate of marble. Wollastonite is somewhat softer than marble and limestone, and a 100-mesh reserve sample was found to contain respective fractions of 78.4 per cent and 60 per cent of <200- and <325-mesh material. The terminus of the single curve for wollastonite dissolution was beyond the right-hand edge of the graph, since 95 per cent of the mineral had been dissolved at the end of 16 hours of extraction. The

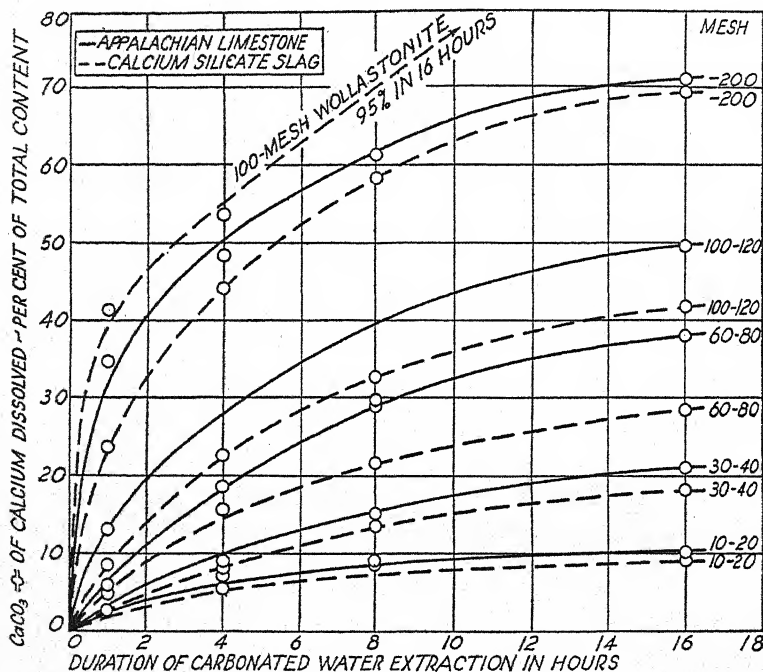


FIG. 2. RELATION OF PARTICLE SIZE TO SPEED OF DISSOLUTION OF CALCIUM SILICATE SLAG IN WATER SATURATED WITH CO_2 AT 5°C .

Slag, limestone, and wollastonite charges, 0.5 gm. per liter; constant agitation

amount of CO_2 requisite for bicarbonate dissolution of the calcium silicate of the wollastonite, however, was approximately twice that requisite for dissolution of the marble. Since the quantity of CO_2 injected into the aqueous suspension of amorphous wollastonite was more than adequate to effect its complete dissolution, a similar potential prevailed in the aqueous suspension of the glassy slag of lower calcium silicate content. This indicates that the density of the slag is the dominant causal factor in its somewhat less rapid dissolution.

The curves of figure 3 show quantities of P_2O_5 and fluorine dissolved progressively in the extracts utilized to obtain the calcium bicarbonate values of

figure 2. A comparison of the curves of figure 2 with those of figure 3 indicates concomitant dissolution of calcium silicate, phosphate, and fluoride. The ultimate percentage extraction of fluorides exceeded that of P_2O_5 from each separate. More than half of the phosphate content, and of the fluoride also, was dissolved from the <200-mesh slag by 16-hour carbonated-water extraction.

POT STUDIES WITH QUENCHED CALCIUM SILICATE SLAG

In the present pot studies, quenched slags from rock phosphate reduction furnaces were compared with limestone treatments, upon the basis of $CaCO_3$ -equivalences. Unless otherwise stated, the two liming materials were of 100-

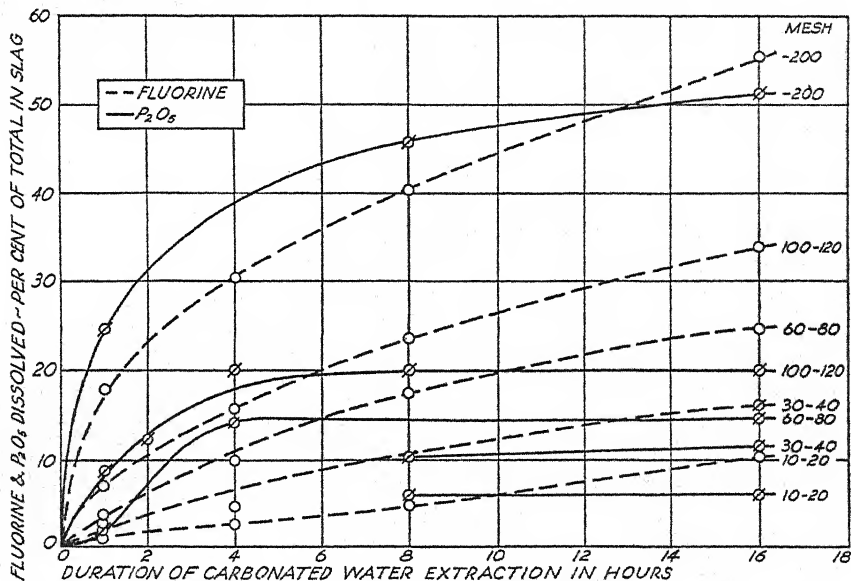


FIG. 3. RELATION OF PARTICLE SIZE TO SPEED OF DISSOLUTION OF FLUORINE AND P_2O_5 CONTENTS OF CALCIUM SILICATE SLAG IN WATER SATURATED WITH CO_2 AT $5^\circ C$.

Slag charge, 0.5 gm. per liter; constant agitation

mesh, and single full depth incorporations were made. Potassium was supplied as K_2SO_4 to all pots, and top dressings of ammonium nitrate were applied to nonlegumes. Liming materials and potassium sulfate were incorporated simultaneously, and the treated soils were aged 3 weeks before any phosphatic treatment. In most cases, three pots were utilized for each treatment, and the upper 2.5-inch soil zone was removed after each harvest and conditioned for the succeeding crop.

Hartsells fine sandy loam

Red clover, sudan grass, red clover, and sudan grass were grown in that succession between July 22, 1938, and September 9, 1939. The three last mentioned crops are shown in plate 2, figure 1.

The superiority of slag S-349 of tables 1 and 2 over limestone is shown by the response of the initial crop of sudan grass to 2-ton and 4-ton additions. Inadequacy of the 1-ton CaCO_3 -equivalent additions of both liming materials is shown by the three successive crops. Slag was more effective than limestone in each comparison of growth of second and third crops, clover and sudan grass, respectively. Because of the relatively short period of aging to induce their reaction with the soil acidoids, the liming materials would not be expected to show their respective maximal beneficial effects upon the initial crop. The effect of aging and of root activities upon dissolution of the liming materials in the soil is shown by greater growth from the second seeding of sudan grass. The progressive benefit from the slag can be attributed to a cumulative liming effect and to additional supplies of P_2O_5 made available to the plants as a consequence of further decomposition of the slag. This relationship was maintained also in related studies, in which limed Hartsells soil was aged and phosphated subsequently with triple superphosphate, fused rock, or calcium metaphosphate.

Dickson silt loam

Plate 2, figure 2, shows the response to equivalent additions of limestone and slag S-349 of tables 1 and 2 on unphosphated Dickson silt loam. Growth was increased with increases in liming rates, and each slag treatment was more effective than its limestone parallel. Apparently, the chief immediate need of this soil is lime, rather than phosphate. The slag was more beneficial in each comparison, but the margin of superiority over limestone was less than that found for other soils.

Somewhat greater effectiveness of the liming materials was shown by the crop of sudan grass grown after opportunity had been afforded for the calcium content of these materials to enter the soil systems. Response from slag exceeded that from limestone also in related studies in which limed and aged Dickson soil was phosphated subsequently with superphosphate, calcium metaphosphate, or fused rock.

Hagerstown silt loam

The response to slag S-349 of tables 1 and 2 at three rates on Hagerstown silt loam is shown in plate 2, figure 3. This well-buffered soil had an initial pH of 5.4 and was more responsive to light liming than were the Hartsells and Dickson soils. Greater response came from slag, for each rate and crop. The effect of aging upon dissolution of limestone and of slag was manifested again by greater yields of sudan grass from its second seeding. This relationship prevailed also in related studies, when slag and limestone were supplemented by subsequent additions of triple superphosphate, fused rock, or calcium metaphosphate, at equivalent rates of 150 pounds and 450 pounds of 16 per cent superphosphate.

Bodine silt loam

The relation of particle size to rate of treatment was considered in trials with Bodine silt loam, obtained from near Wilson Dam, Alabama. Conventional incorporations of 35-mesh screenings of slag S-686 of tables 1 and 2 were compared with heavier incorporations of unground slag of 4-mesh, which contained only 8 per cent of material finer than 35-mesh. Plate 2, figure 4, shows the first three of four successive crops—red clover, sudan grass, red clover, and carrots. Meager yields from N and K treatments, with and without 35-mesh limestone at the 3-ton rate, were in marked contrast to heavy yields induced by equivalent additions of slag of like fineness. Response from the coarse slag at the rate of 5 tons was somewhat greater than that from the 3-ton rate. Response from the 35-mesh slag was greater also for the 5-ton incorporation. It is evident that this soil requires something that is present in the slag and absent from the limestone.

Paden silt loam

Fertilizer requirements of Paden silt loam are not well established. This brownish gray acidic soil from Humphreys County, Tennessee, is considered representative of a group of well-drained hardpan soils of the uplands and bench lands of that part of the Tennessee River Watershed within the Highland Rim in West Tennessee.

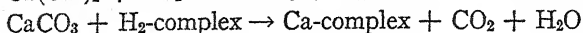
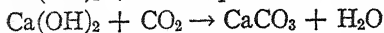
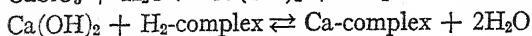
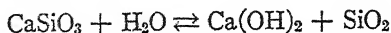
The initial pH of the Paden soil was 5.2, and 61 per cent of its saturation capacity was accounted for by Ca and Mg. Elevation of its pH to 6.8 was effected by 2 tons of 100-mesh limestone, and the same change was induced by 3 tons of 35-mesh slag. Unground quenched material, S-653 of table 1, was compared with 3-ton additions of 20-mesh and 35-mesh slag and 35-mesh limestone and with both meshes of slag at the rate of 5 tons.

Successive crops of sudan grass, sweet clover, and sudan grass, grown between June 6 and March 28, 1940, are shown in plate 2, figure 5. The response to each of the five slag treatments exceeded by far the response to an equivalent addition of limestone. Although the 5-ton addition of coarse slag proved decidedly more effective than the 3-ton addition of 35-mesh limestone, greater beneficial effects of 3-ton and 5-ton additions of both 20- and 35-mesh slag demonstrate that the regular quenched product, when used at ordinary rates, is not sufficiently fine to induce maximal early response. The aggregate crop response from slag of 20- and of 35-mesh was three times that induced by 35-mesh limestone at the 3-ton rate. Somewhat comparable response came from the 20- and 35-mesh slag separates.

DISCUSSION OF BENEFICIAL EFFECTS OF QUENCHED SLAG

The responses illustrated in plate 2 are typical of those found in similar trials with other soils and demonstrate the superiority of slag over equivalent high-calcic limestone of like particle size at rates up to 5 tons. The calcium silicate

content of the slag may be considered as a source of a continuous supply of calcium hydroxide that enters the soil complex, either directly or through carbonate forms:



It cannot be concluded unqualifiedly, however, that the calcium silicate content of the slag proved superior to an equivalence of limestone. Spectrographic examinations indicate that the slag carries substantially all of the original rock phosphate components, except phosphorus. The slag therefore supplies minor elements, as well as solvated silica, not supplied by the limestone.

Quenched slag shows to best advantage when added to phosphorus-deficient soil, and the response is similar to that induced by limestone plus phosphate. The slag has a relatively low P_2O_5 content, but incorporations of 2 to 5 tons of slag of 70 per cent CaCO_3 -equivalence introduce approximately 60 to 150 pounds of P_2O_5 , about one-half of which is registered as "available" by a single digestion of a 1-gm. charge with the conventional ammonium citrate solvent. Moreover, the relatively small quantity of P_2O_5 is distributed uniformly throughout a large bulk of calcium silicate. Concomitant dissolution of the phosphate and solvation of silica occur when the calcium silicate body of the slag is dissolved. That part of the phosphate not dissolved during the dissolution of the silicate is in a state of division far beyond that possible by pulverization of rock phosphate. The solubility of pulverized apatite cannot be taken as a measure of the solubility of the phosphate content of the slag, although the high ratio of calcium fluoride to calcium phosphate in the slag indicates that the P_2O_5 content is most probably in the form of fluorophosphate.

It is concluded, therefore, that the beneficial effect of quenched slag is not due solely to its effectiveness as a liming material. Supplemental comparisons between limestone and calcium silicate, both mineral and precipitated forms, with and without phosphates, did not warrant the conclusion that beneficial effects are due either to "secondary" elements carried by the starting rock phosphate or to solvated silica derived from the slag. The extent of superiority of an incorporation of calcium in the form of slag, over an equivalence supplied by either limestone or wollastonite, is attributed, therefore, primarily to the associated content of basic phosphate. When the calcium silicate framework of the slag undergoes dissolution, the phosphate content of the slag becomes disseminated throughout the soil as colloidal material of relatively high solubility.

POSSIBLE DELETERIOUS EFFECT OF QUENCHED SLAG

Observations and findings

In initial comparisons of limestone and quenched slag in the pot studies, 100-mesh materials only were used. A question arose as to the economy of using

unground slag at heavy rates in lieu of finer materials at conventional rates. It was postulated that ultimate toxicity might come from repetitions of slagging with coarse material at rates required to induce early response by crops, and that any such toxicity would be expedited by heavy single incorporations of finely ground slag. Full-depth incorporations of slag S-686 of table 1, therefore, were made to Montevallo silt loam at rates of 5, 10, 20, and 40 tons of 20-60-mesh and 100-mesh material, in parallel with limestone. The soils were allowed to age 3 weeks between incorporations and seeding.

Yields of the sudan grass and also those of a preceding crop of sweet clover and final pH values are given with legend for the sudan grass photographs of plate 2, figure 6. The results show superiority of slag over limestone for 100-mesh material at the rate of 5 tons and superiority also for the 20-60-mesh slag at heavier rates. For the first time, however, definite and even lethal toxicity was induced by 100-mesh quenched slag and without corresponding effect from limestone. The heavy rates of 20-60-mesh slag did not develop the toxicity induced by the heavy incorporations of 100-mesh slag. The response from the 40-ton addition of 100-mesh limestone actually exceeded that from any of the smaller additions of limestone of either 100-mesh or 20-60-mesh.

Discussion

The scope of the present paper does not admit of detailed consideration of causes for the toxicity induced immediately by the heavy incorporations of ground slag. Those causes and the duration of toxicity are objectives of further study, but certain pertinent observations will be noted briefly as to potential alkalinity of the slag and its concentration of fluorides. These factors seemed probabilities for the toxicity induced by heavy incorporations of the finely ground slag.

Data of table 2 show that the aqueous extracts of the quenched slag are strongly alkaline. Liming treatments induced pH values ranging from 7.2 to 8.7 in the Montevallo soil. In each comparison, save for the minimal rate, pH induced by slag exceeded that induced by an equivalence of limestone. Ordinarily, elevation in pH brings decreases in availability of K and P. The effect upon K solubility was minimized, however, by a substantial supplement of potassium sulfate, and sodium acetate extraction registered a high content of available P_2O_5 for each slagged soil.

Analyses of table 1 show that the medial calcium fluoride content of seven slags was 5.2 per cent. The rate of fluoride removal by rainwater leachates from heavily slagged soil was an undetermined factor, although interpretation of results from a previous lysimeter study (11) points to the conclusion that concentration of calcium salts resultant from dissolution of the slag would have caused a repressive effect upon removal of fluorides, if the undrained pots had been subjected to leaching.

Analyses of several crops demonstrated that increased rates of slagging did not bring about marked or consistent increases in uptake of fluorine. Moreover, yields of sudan grass gave no indication of fluoride-induced toxicity from

20-ton additions of 100-mesh wollastonite supplemented with sufficient triple superphosphate to supply the amount of calcium fluoride carried by the 20-ton addition of 100-mesh slag. Yields from joint incorporation of wollastonite and triple superphosphate were virtually the same as those obtained when the wollastonite was supplemented likewise by fluoride-free monocalcium phosphate. The acidity of the two phosphatic supplements was sufficient, however, to neutralize immediately 52 per cent of the potential alkalinity of the 20-ton addition of wollastonite. Hence, in these two control systems, the potential alkalinity of the wollastonite treatment was decreased to 10-ton CaCO_3 -equivalence, with and without the amount of fluorides carried by an equivalent amount of quenched slag.

A possible explanation is that heavily slagged soil is subjected to the continuous action of $\text{Ca}(\text{OH})_2$ during dissolution of the slag. The calcium silicate of heavy incorporations of the slag undergoes hydrolysis to produce calcium hydroxide in quantities capable of depleting the soil of its normal CO_2 content for protracted periods, during which substantial diminution, or even inhibition, of aerobic bacterial activity occurs. Because of this diminution of CO_2 of soil origin, and with limited supplies of CO_2 from the aerial atmosphere, carbonation of the liberated hydroxide probably does not keep pace with the liberation of $\text{Ca}(\text{OH})_2$. Even though some quantities of calcium carbonate may be formed in the soil, as an initial effect of slagging, further transition into bicarbonate is precluded as long as excessive quantities of hydrolyzable CaSiO_3 persist in the slagged soil. Heavy incorporations of slag therefore produce an effect analogous to that to be expected from a continuous supply of solute $\text{Ca}(\text{OH})_2$ into the soil during a protracted period.

Dissolutions of equivalent quantities of limestone and slag in the soil do not develop systems of identical immediate alkalinity. Disintegration of each mol of CaCO_3 of limestone by soil acidoids results in *liberation* of a mol of CO_2 , and the soil system tends to buffered conditions of equilibrium, whereas transition of each mol of calcium silicate into CaCO_3 *removes* one mol of gaseous CO_2 from the soil system. Moreover, only one mol of CO_2 is required to effect bicarbonate solution of limestone, whereas two mols are utilized for transition of one mol of calcium silicate into $\text{CaH}_2(\text{CO}_3)_2$.

Dissolution of moderate incorporations of finely ground slag probably does not diminish the CO_2 of the soil atmosphere with such rapidity as to induce alkalinity harmful to biochemical activities. Amounts of CO_2 supplied by soil and aerial atmospheres probably are adequate to prevent detrimental disruption of a favorable balance between the calcic solids and the gaseous phase of CO_2 in the soil.

This postulation may be only one factor contributive to the toxicity induced by heavy incorporations of 100-mesh material and not by 20-60-mesh slag. Repression in the availability of minor elements, complementary dissolution of sodium and potassium silicate components, and effects of sulfides and phosphides are also possible factors, even though the slag contains relatively small percentages of these two reduction products.

SUMMARY AND CONCLUSIONS

The primary objective of the present study was to determine the effectiveness of quenched calcium silicate slag as a liming material. Quenched slag from rock phosphate reduction furnaces is essentially a brittle, glassy, calcium silicate, formed chiefly by replacement of substantially all of the P_2O_5 content of the raw rock by additive silica. It would be confusing and erroneous to use the well-established term "basic slag" in referring to this relatively new by-product, characterized by high-silicate and low-phosphate content.

Speed of dissolution of quenched slag of variant particle size in carbonated water and components of the resultant extracts were determined. Solubility of ground quenched slag proved to be comparable to solubility of limestone of similar fineness.

Evaluated by response of leguminous and nonleguminous crops on six soils, 100-mesh slag invariably proved superior to limestone at ordinary rates. Similar superiority was shown by heavy additions of coarse slag. Quenched slag also invariably proved superior to equivalent quantities of other forms of calcium silicate. Plant response to the quenched slag was particularly good on phosphorus-deficient soils and was comparable to response obtained from slag and limestone plus acidic phosphates. It is concluded that, at liming rates, the slag supplies beneficial amounts of P_2O_5 .

Disintegration of the slag grains probably results in dispersion of colloidal basic phosphate possessing a solubility rate far beyond that found for laboratory-prepared and natural fluorophosphates. It is believed, therefore, that superiority of the slag over equivalent quantities of carbonates and silicates is due primarily to the substantial amounts of P_2O_5 supplied to the soil when the slag is used at rates in keeping with its effectiveness as a liming material.

The possibility that initially beneficial heavy additions of coarse material, and repetitions of ordinary additions, might induce ultimate toxicity upon plant growth was considered. To expedite the advent of such possible effect, incorporations of 20-60-mesh and of 100-mesh slag were made at rates of 5, 10, 20, and 40 tons. No deleterious effect was induced by even the 40-ton incorporation of 20-60-mesh material, but definite toxicity was induced on the first two crops, sweet clover and sudan grass, by the 100-mesh material at rates above 10 tons. Progressive supply of $Ca(OH)_2$ from the heavy incorporations and resultant excessive alkalinity and depletion of CO_2 in the soil system is suggested as an explanation of this toxicity.

The findings point to the conclusion that slag of particle size between 4-mesh and 35-mesh can be used safely and advantageously at rates up to 10-ton $CaCO_3$ -equivalence per 2,000,000 pounds of soil, and that 2- to 3-ton $CaCO_3$ -equivalence of material passing a 20-mesh sieve constitutes a safe and effective liming for ordinary soil depth.

The ultimate effects of heavy single incorporations, and the cumulative effects of repetitions of ordinary additions, upon soil constituents and plant composition, are being studied.

REFERENCES

- (1) AMES, J. W. 1916 Blast-furnace slag as a source of bases for acid soils. *Ohio Agr. Exp. Sta. Mo. Bul.* 1: 359-62.
- (2) BARNETTE, R. M. 1924, 1926 Synthetic calcium silicates as a source of agricultural lime: I, II, III. *Soil Sci.* 18: 479-491 (1924); 21: 443-453 (1926); 22: 459-466 (1926).
- (3) COOK, H. L., AND CONNOR, S. D. 1936 A study of the basicity of dolomite, rock phosphate, and other materials in preparing non-acid-forming fertilizers. *Jour. Amer. Soc. Agron.* 28: 843-855.
- (4) COWLES, A. H. 1917 Calcium silicates as fertilizers. *Chem. and Metall. Engin.* 17: 664-665.
- (5) COWLES, A. H. 1917 Soluble Silica as an Essential Fertilizer. (Addendum by Scheidt, A. W.). The Electric Smelting and Aluminum Company, Sewaren, N. J.
- (6) HARTWELL, B. L., AND PEMBER, F. R. 1920 The effect of dicalcium silicate on an acid soil. *Soil Sci.* 10: 57-60.
- (7) JOHNSTON, J. F. W. 1864 Lectures on the Applications of Chemistry and Geology to Agriculture, New Ed. with App. C. M. Saxton, New York.
- (8) MACINTIRE, W. H. 1916 Factors influencing the lime and magnesia requirements of soils. *Tenn. Agr. Exp. Sta. Bul.* 115.
- (9) MACINTIRE, W. H., ET AL. 1934 The conservation of burnt lime, limestone, dolomite, and calcium silicate in soil, as influenced by methods of incorporation. *Tenn. Agr. Exp. Sta. Bul.* 152 and *Va. Agr. Exp. Sta. Tech. Bul.* 54.
- (10) MACINTIRE, W. H., HARDIN, L. J., AND OLDHAM, F. D. 1936 Properties of quenched and unquenched slags and effects of their admixtures with phosphatic fertilizers. *Indus. and Engin. Chem.* 28: 48-57.
- (11) MACINTIRE, W. H., SHAW, W. M., AND ROBINSON, B. 1936 A barium-fluorine study: The fate of added BaSiF_6 and its effect upon sulfates and other soil components. *Tenn. Agr. Exp. Sta. Bul.* 155.
- (12) MACINTIRE, W. H., AND WILLIS, L. G. 1914 A comparison of silicates and carbonates as sources of lime and magnesia for plants. *Jour. Indus. and Engin. Chem.* 6: 1005-1008.
- (13) MIDGLEY, A. R. 1932 Overliming acid soil. *Jour. Amer. Soc. Agron.* 24: 822-35.
- (14) SCHOLLENBERGER, C. J. 1922 Silica and silicates in relation to plant growth and composition. *Soil Sci.* 14: 347-62.
- (15) SHEDD, O. M. 1922 Effect of certain calcium compounds and other substances on the yield and calcium content of some crops. *Soil Sci.* 14: 233-46.
- (16) THOMAS, W. 1930 The effect of colloidal silica on the absorption of phosphoric acid by plants. *Science* 71: 422-3.
- (17) WEISER, V. L. 1933 Fixation and penetration of phosphates in Vermont soils. *Vt. Agr. Exp. Sta. Bul.* 356.
- (18) WHITE, J. W. 1928 The agricultural value of specially prepared blast furnace slag. *Pa. Agr. Exp. Sta. Bul.* 220.
- (19) WIANCKO, A. T., WALKER, G. P., AND CONNER, S. D. 1929 Comparison of soil liming materials. *Ind. Agr. Exp. Sta. Bul.* 329.
- (20) WILLARD, H. H., AND WINTER, O. B. 1933 Volumetric method for determination of fluorine. *Indus. and Engin. Chem., Analyt. Ed.* 5: 7.

PLATES

PLATE 1

MICROPHOTOGRAPHS OF CALCIUM SILICATE SLAGS OF TABLE 1 FROM ROCK PHOSPHATE
REDUCTION FURNACE OPERATIONS AT THREE PLANTSMagnification $\times 80$

- FIG. 1. Wilson Dam, S-348, unquenched.
- FIG. 2. Wilson Dam, S-349, quenched in water.
- FIG. 3. Wilson Dam, S-686, quenched by stream of water.
- FIG. 4. Tennessee commercial product, S-657, quenched by stream of water.
- FIG. 5. Florida commercial product, S-791, unquenched.
- FIG. 6. Florida commercial product, S-792, quenched in water.



FIG. 1

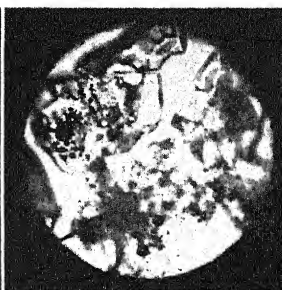


FIG. 2



FIG. 3

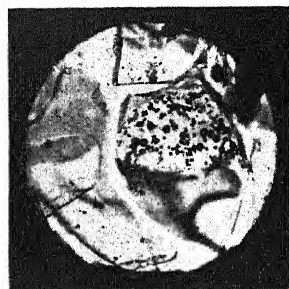


FIG. 4

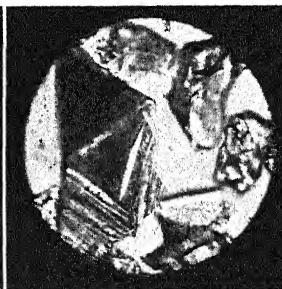


FIG. 5

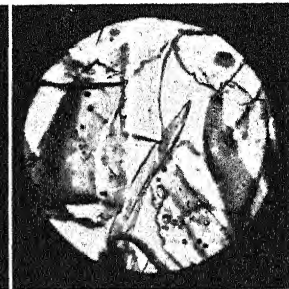


FIG. 6

SEVERAL SOILS

FIGS. 1, 2, 3. Top to bottom, successive crops of sudan grass, red clover, sudan grass.

TREATMENT* (LEFT TO RIGHT)	TOTAL YIELDS, THREE CROPS†		
	Hartsells fine sandy loam (fig. 1)	Dickson silt loam (fig. 2)	Hagerstown silt loam (fig. 3)
Unlimed‡	25.6
Limestone, § 1 ton	6.6	31.2	30.5
Slag, § 1 ton	21.7	38.1	45.2
Limestone, 2 tons	20.7	39.7	38.7
Slag, 2 tons	38.4	40.6	53.4
Limestone, 4 tons	35.0	48.6	47.5
Slag, 4 tons	74.2	62.4	60.1

* All pots received potassium sulfate, rate of 300 pounds of K_2O , incorporated and aged 3 weeks with liming materials, and sufficient N as ammonium nitrate to assure optimal growth of sudan grass.

‡ Dry weight, mean grams per pot. † Unlimed pots not shown in figures 1 and 2.

§ Liming materials, 100-mesh; slag S-349 of table 1; rates based on $CaCO_3$ per 2,000,000 pounds of soil.

FIG. 4. *Bodine silt loam*. Top to bottom, successive crops of red clover, sudan grass, red clover.

TREATMENT* (LEFT TO RIGHT)	TOTAL YIELDS, THREE CROPS†
Unlimed	4.6
Limestone, ‡ 35-mesh, 3 tons	5.3
Slag, ‡§ 35-mesh, 3 tons	33.8
Slag, ‡ 4-mesh, 3 tons	13.2
Slag, 35-mesh, 5 tons	43.2
Slag, 4-mesh, 5 tons	18.4

* See * figures 1, 2, 3; † see † figures 1, 2, 3.

‡ Liming based upon $CaCO_3$ per 2,000,000 pounds of soil.

§ All slag treatments were S-686 of table 1.

|| Contained 8 per cent of material finer than 35-mesh.

FIG. 5. *Paden silt loam*. Top to bottom, successive crops of sudan grass, sweet clover, sudan grass.

TREATMENT* (LEFT TO RIGHT)	TOTAL YIELDS, THREE CROPS†
Unlimed	14.1
Slag, ‡ 4-mesh, § 5 tons	47.2
Slag, 20-mesh, 3 tons	58.7
Limestone, 35-mesh, 3 tons	28.7
Slag, 35-mesh, 3 tons	61.7
Slag, 20-mesh, 5 tons	78.0
Slag, 35-mesh, 5 tons	87.1

* All pots received potassium sulfate, rate of 130 pounds K_2O , incorporated in upper 3 inches, and sufficient N as ammonium nitrate to assure optimal growth of sudan grass.

† See † figures 1, 2, 3.

‡ All slag treatments were S-653 of table 1; incorporations aged 3 weeks before seeding.

§ Contained 13.5 per cent of material finer than 35-mesh.

FIG. 6. *Monterallo silt loam*.

TREATMENTS* (LEFT TO RIGHT)		YIELDS, GRAMS DRY WEIGHT, PER POT†			
		Final pH‡	Sweet clover§	Sudan grass	Aggregate
Top row	Control	6.2	0.33	2.0	2.33
	Slag, 100-mesh, 5 tons	7.2	5.10	27.0	32.10
	Limestone, 100-mesh, 5 tons	7.4	1.36	10.1	11.46
	Slag, 20-60-mesh, 10 tons	8.1	4.37	22.3	26.67
	Slag, 100-mesh, 10 tons	8.5	3.27	25.0	28.27
	Slag, 20-60-mesh, 20 tons	8.2	4.44	23.8	28.24
	Slag, 100-mesh, 20 tons	8.5	0	0.8	0.80
Bottom row	Limestone, 20-60-mesh, 20 tons	7.4	1.35	9.0	10.35
	Limestone, 100-mesh, 20 tons	7.7	1.94	13.7	15.64
	Slag, 20-60-mesh, 40 tons	8.4	3.50	19.9	23.40
	Slag, 100-mesh, 40 tons	8.7	0	0.7	0.70
	Limestone, 20-60-mesh, 40 tons	7.6	2.35	12.8	15.15
	Limestone, 100-mesh, 40 tons	7.6	2.20	14.4	16.60

* All pots, including control, received potassium sulfate, rate of 200 pounds of K_2O , incorporated and aged 3 weeks with liming materials, and sufficient N as ammonium nitrate to assure optimal growth of sudan grass. Liming on basis of $CaCO_3$ -equivalence. Slag separations were obtained by sizing slag S-686 of table 1.

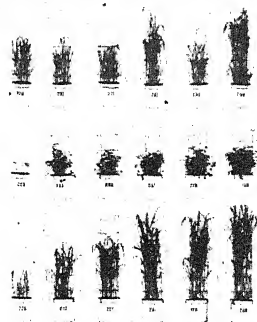


FIG. 1

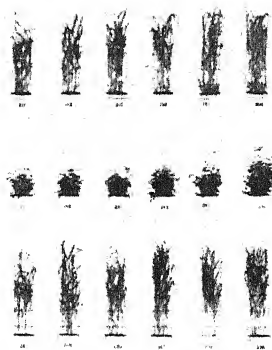


FIG. 2

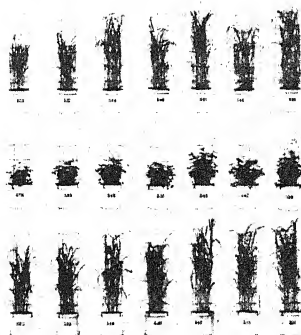


FIG. 3

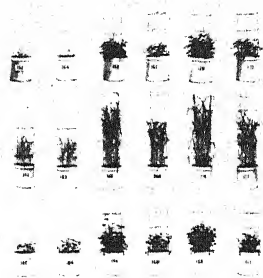


FIG. 4

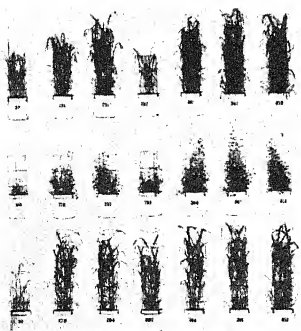


FIG. 5

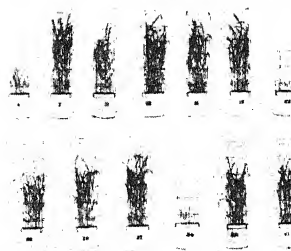
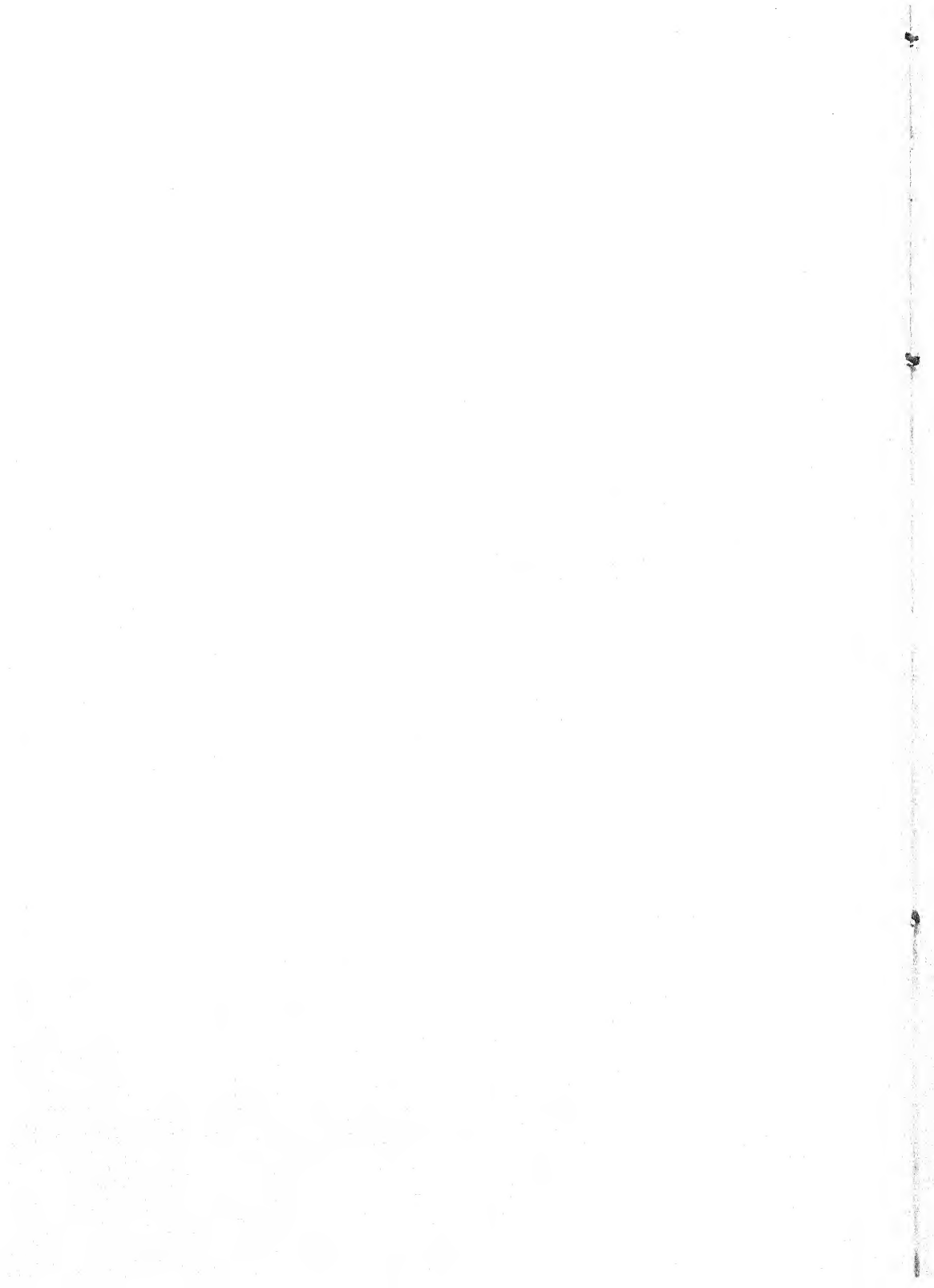


FIG. 6



THE USE OF SOIL MOISTURE CHARACTERISTICS IN SOIL STUDIES

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In recent years considerable activity has been displayed in the investigation of the relationship between the moisture content of soil and the pressure deficiency, or suction, with which that moisture is held, and we are now familiar with curves obtained by plotting these two mutually dependent variables against each other. Reference to this work in discussion is, however, rendered difficult by the fact that such curves have no generally accepted name; we can compare the situation with that which would exist in the field of thermionic valve, or vacuum tube, technology if we could not refer briefly to "valve characteristics," and to the difficulty of discussion of heat engines if we had no general acceptance of the term "indicator diagram." In order to remove the difficulty, the writer proposed the name "soil moisture characteristic curve" at the Conference of the First Commission of the International Society of Soil Science, held at Bangor, Wales, in the summer of 1939.¹ This name may conveniently be contracted to "soil moisture characteristic" for general use. This name is equally applicable and descriptive, whether the moisture content is plotted against the pressure deficiency itself or whether some convenient function of the latter, such as the logarithm, is used, as is sometimes advantageous.

Though soil moisture characteristics are of interest in themselves, it does not seem to be generally realized that they are capable of a much wider use and that they provide a method of investigation in soil studies. In this paper we propose to demonstrate the method by applying it to the study of tilth and soil stability. Use of soil moisture characteristics in this way depends on the fact that these characteristics may be interpreted as showing the pore size distribution within the soil (2, 4, 7, 10). Thus they play a part analogous and complementary to mechanical analysis: they give the same sort of information about the pores as that given by mechanical analysis about the particles.

The pressure deficiency p required to maintain soil at a given moisture content is the pressure difference between the two sides of the air-water interface within the soil pores. If r is the radius of curvature of a spherical interface in equilibrium at this pressure difference, then

$$p = 2s/r \quad (A)$$

where s is the surface tension. Pores into which the interface may retreat via channels of radius greater than r will, at this pressure deficiency, be emptied

¹Publication of the transactions of this conference is temporarily postponed.

of all water except that which remains in nooks and crannies in such small quantities as can be contained within an interface of smaller radius of curvature. Pores into which the interface cannot retreat except through channels of radius smaller than r will remain full of water. As the suction pressure increases, the moisture content progressively decreases as a result of the successive emptying of pores of smaller and smaller radius. The pressure axis (x coordinate) of the moisture characteristic may thus be regraduated in terms of pore size by means of equation (A). The y coordinate of any point on the moisture characteristic then gives us the quantity of water held in pores of radius smaller than that given by the x coordinate. The shape of the whole characteristic therefore tells us the distribution of the total pore space among pores of different effective sizes. Since the pores are, of course, very irregular in shape, the interfaces are, in general, not truly spherical, and therefore the effective pore size given by equation (A) bears a relation to the true pore size which is of the same nature as the relation between effective and true particle size in mechanical analysis. The moisture characteristic method of analysis has the great advantage that the experimental technic involves very little violence to the soil sample. This renders it very suitable for the study of the state of aggregation of soil in the natural field condition and for the investigation of soil stability, that is, ability to withstand natural disintegrating forces. Any method of aggregate analysis, whether by sifting or by mechanical analysis without dispersion, entails such a measure of violence that the results are open to the criticism that the size distribution of the aggregates thus found is due to the process of analysis and may not have existed in the soil before analysis.

DESCRIPTION OF THE METHOD

It is possible, as a preliminary, to demonstrate the validity of the assumption that soil moisture characteristics indicate the pore size distribution in certain cases. If we take a crumb fraction of a clay known to be very stable, such as Gault, and a similar sand fraction, it may reasonably be expected that the size distribution of the intercrumb pores should be similar to the pore size distribution in the sand. To show this, a crumb fraction of Gault clay was separated by sifting in the dry state, the clay clod being first crushed as gently as possible with a wooden pestle. The crumb fraction chosen was that which passed the 2-mm. sieve and was retained on the 1-mm. sieve. The same sieves were used to separate a sand fraction. The moisture characteristics of the two materials were obtained by Haines' Büchner funnel method (4). (In the whole of the work described here, it will be seen that great pressure deficiencies were not required, and the method of Haines was used throughout.) Care was taken to ensure the same degree of packing, as nearly as possible, in the two cases by tapping down the sand and crumbs as tightly as could be done without risk of disintegration of the latter. Curve *a*, figure 1, shows the moisture characteristic of the sand fraction, and curve *b*, that of the crumb fraction, after a correction necessary for a true comparison. The necessity for this correction

arises because the crumbs swell when saturated prior to the taking of the moisture characteristic, and the size distribution of the aggregates, and therefore of the intercrumb pores, is no longer similar to that in the sand. If we assume that the swelling merely alters the linear dimensions equally in all directions, without changing the shapes of the crumbs and pores, then we can see from equation (A) that for a given percentage moisture content the pressure deficiency varies in inverse proportion to the linear dimensions. The linear dimensions of the clay crumbs were found to increase in the ratio 1.08 to 1, and therefore all measured pressure deficiencies were multiplied by 1.08 in order to arrive at the pressure deficiency which would have obtained in the absence of swelling.

The similarity between the two characteristics is at once apparent. The chief difference is that the moisture content of the clay is at all pressure deficiencies greater than the sand moisture content; this difference is due, of course, to the water held in the crumbs themselves, as distinct from that contained in the pores between them. Apart from this difference, there is no reason why the two characteristics should be identical, since there is no reason why the size distribution of particles of two quite different materials, even in such a restricted range of sizes, should be identical, and this distribution, together with the shapes and packing of the particles, determines the pore size distribution. There is, however, reason to expect close similarity, and this is, in fact, observed. The point which emerges is that the moisture characteristic of the clay crumbs indicates the presence of a group of large pores which may be identified with the intercrumb pores, on account of their similarity of size distribution to the sand pores. This group of pores is shown more clearly in figure 2, obtained by differentiating the curves of figure 1 and plotting the absolute values of the slopes. In these curves the area enclosed between the ordinates p and $p + \delta p$ gives the percentage space occupied by pores of such size that they are emptied of water by an increase of suction from p to $p + \delta p$. The bulk of the pores in the sand occurs about an effective pore radius of 0.022 cm., while for the clay crumbs the corresponding prevalent radius, corrected to dryness in the manner already described, is 0.025 cm. The moisture characteristic of the clay has not been extended to great suction pressures; such a procedure would give us the size distribution of a second group of pores, much smaller in effective radius, within the clay crumbs and proper to the clay itself. Such a moisture characteristic, which would be similar to some of those published by other workers, has no application to the work described in the present paper. It is possible, of course, that if we took a soil sample in the field condition, or whole soil instead of a crumb fraction of limited size range, two pore groups would not be distinguishable. This could occur if the crumbs were to fit closely together, as happens in many clay subsoils, giving negligible pore space; or it could be caused by the intercrumb pore space tailing imperceptibly into intracrumb pore space. In such cases it would be quite unjustifiable to speak of distinct groups of pores.

It will be observed that we have confined our description of the soil conditions to those features which are clearly indicated by the moisture characteristics; we have drawn attention to groups of pores where such are demonstrably present, but have particularly avoided giving them misleading names, such as "capillary" or "noncapillary pore space," which imply a knowledge of their physical properties that we do not possess and which, in fact, are not capable

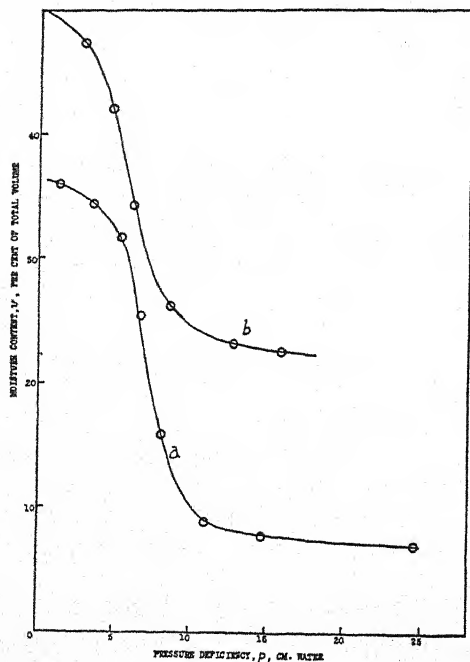


FIG. 1

FIG. 1. MOISTURE CHARACTERISTICS OF (a) SAND FRACTION, GRAIN SIZE BETWEEN 1 AND 2 MM., (b) GAULT CLAY CRUMB FRACTION OF SIMILAR SIZE RANGE

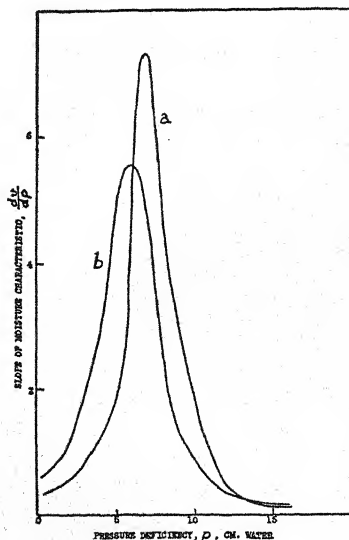


FIG. 2

FIG. 2. DIFFERENTIAL CURVES DERIVED FROM FIGURE 1, SHOWING PORE SIZE DISTRIBUTION IN (a) SAND FRACTION AND (b) GAULT CLAY CRUMB FRACTION

(a) Marked peak at a pressure deficiency of 7.0 cm. water indicates particular abundance of pores of radius grouped about 0.022 cm.

(b) Marked peak at a pressure deficiency of 6.0 cm. water indicates particular abundance of pores of radius grouped about 0.025 cm.

of precise meaning. The application of this apparently artificial case will be apparent later.

STUDY OF UNDISTURBED FIELD SAMPLES

The nature of the pore space of soil in the field, undisturbed either by sampling operations or by experimental methods, is of importance in drainage investigations in heavy clays. The wax impregnation method used by Nicholson (9) in such studies is of value in giving a qualitative comparison of the pore

space at various depths or in various circumstances of tillage, but the drying of the soil in the process causes shrinkage which much exaggerates the pore volume, including soil cracks. The moisture characteristic method of analysis fills the gap by giving a quantitative comparison, and is thus complementary to the wax impregnation method.

The series of moisture characteristics shown in figure 3 was obtained by using soil samples from various depths in old grassland on Oxford clay. These samples were obtained in metal cylinders about 5.5 cm. in diameter and 2.5 cm. deep. The soil was first removed to a depth corresponding to the top of the required sample, and the exposed surface was pared flat. The sampling ring was then pressed in until the top was flush with this prepared surface, and the sample was removed in its ring, care being taken to remove plenty of spare soil with it. The excess soil was removed in the laboratory, crumb by crumb, until the lower surface was level with the bottom of the containing ring and in an "unpuddled" condition. The sample was then pressed on the filter paper in the Büchner funnel to ensure reasonable contact. The water level in the funnel was adjusted to the level of the top of the sample, which was left to soak for 24 hours. The excess free water was then drained off and the moisture characteristic taken in the usual way by increasing the suction pressure in stages.

The curves of figure 3 again show the presence of groups of coarse pores, most prominent in the uppermost "tilthy" soil and becoming less marked with increasing depth. In the top inch there appear to be two groups, one distributed about a pore size of 0.072 cm. and one about a pore size of 0.009 cm. It is tempting to identify the first with cracks, root holes, etc., and the second with intercrumb pore space, since this second group bears a resemblance, as will be seen later, to the degenerated pore space of a weathered crumb fraction. This is, of course, pure speculation. At a depth of 2 to 3 inches these groups are much modified, although occurring at about the same pore size. At greater depths it cannot be said with certainty that the groups exist at all. This does not necessarily mean that aggregates do not exist at depth. This soil is one of those mentioned earlier, in which the subsoil crumbs are of an angular, faceted, semicrystalline appearance, fitting closely together in a three-dimensional mosaic, and leaving very few large pores. Thus we see that although the method of moisture characteristics has the advantage of avoiding soil disturbance, it is complementary to, and not a preferable alternative to, aggregate analysis, which has not this advantage. In this case aggregate analysis would give the required information about the presence of aggregates, although possibly in distorted form, whereas the moisture characteristics show, by the absence of large pores, the packing arrangement.

A series of characteristics, as in figure 3, has another possible use. From it can be calculated the amount of drainable water in the soil at any given time, the only additional knowledge required being the moisture profile obtaining at the time. Consider a soil sample at a height of x cm. above the level of the

drains. These drains will cease to remove subsoil water when the water table falls to their level, and the soil sample under consideration will then be in equilibrium with a free water surface x cm. below it, that is, the soil water will have a pressure deficiency of x cm. of water. We can find, from the moisture characteristic of the soil at the depth of the sample, the moisture content at this suction pressure, and all water which at any time is in excess of this is

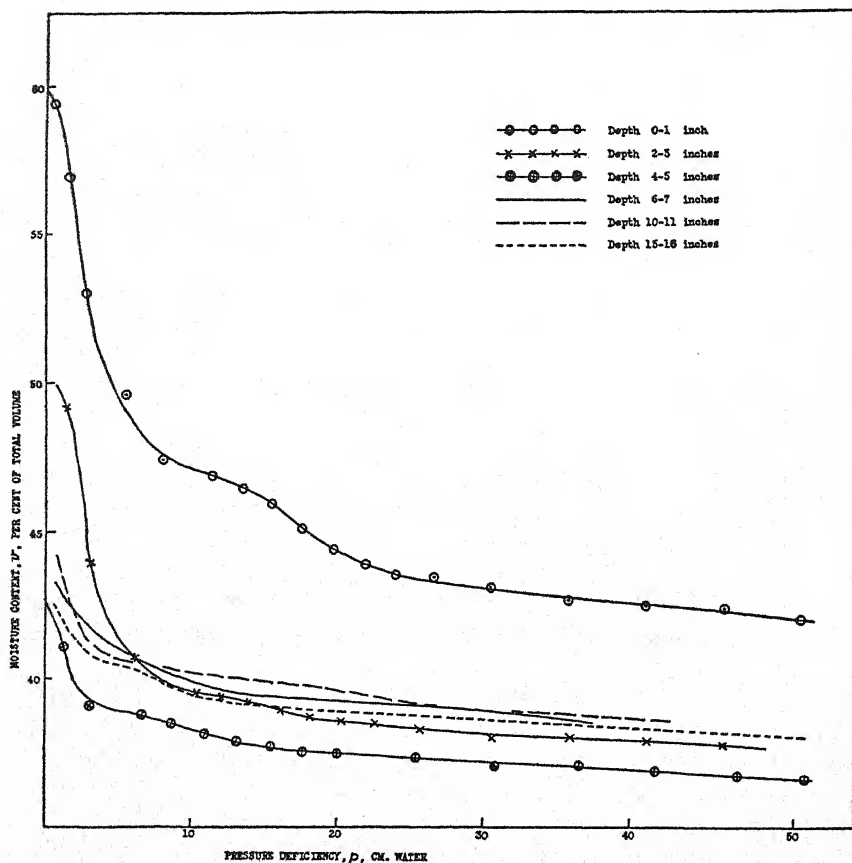


FIG. 3. MOISTURE CHARACTERISTICS OF OXFORD CLAY IN FIELD CONDITION

Experimental points omitted on curves for samples from depths of 6-7, 10-11, and 15-16 inches, to avoid confusion.

drainable. Summation of this excess throughout the soil profile above the drain level gives the total content of drainable soil water. It may be found that the soil water does not reach the drainable value, even when the drains are observed to be running. The inference then is that the drainage water is not, properly speaking, soil water at all but run-off water (not necessarily at the surface) which the crumbs cannot absorb with sufficient speed. Given

time, the crumbs could absorb more water, which, once absorbed, would not again be given up to the drains. This behavior would be observed, of course, only in heavy soils.

STUDY OF SOIL STABILITY

It is a feature of many clay soils of poor calcium status that the clods left by the plough fail to weather properly during the alternate wetting and drying of winter and early spring. Instead of breaking down into a mass of small crumbs, they become rounded and "capped" masses with no visible traces of structure. The process of disintegration of crumbs due to the wetting and drying cycle may be followed very easily by means of a series of moisture characteristics taken on the soil in its original condition and after one or more cycles of wetting (with distilled water) and drying. Such a study was made on a crumb fraction separated by the 2-mm. and 1-mm. sieves as already described. The moisture characteristic of these crumbs shows the presence of the group of coarse pores in all but the most unstable soils, which might slake down at the first wetting after being placed in the Büchner funnel. Another and equal quantity of the same crumb sample was soaked in distilled water for 24 hours and then allowed to become air dry before being placed in the Büchner funnel and saturated in preparation for the taking of the moisture characteristic. Yet another equal quantity was carried through two wetting and drying cycles before the moisture characteristic was taken. This process was continued until all the information required was obtained. The modification of the group of coarse pores at each stage could thus be followed. Here again it may be remarked that almost any process of aggregate analysis would fail to show this process of "weathering" on the score that such analysis involves a disintegration comparable with that due to the weathering under investigation.

The curves of figures 4 and 4a show the course of "weathering" of a sample of Gault clay which, containing ample free calcium carbonate, has a good lime status. Curve 1 again shows the prominent group of intercrumb pores, distributed about a peak at a pore radius of 0.023 cm. After one weathering cycle this group is still prominent, although less so than before, and is now distributed about a peak at the smaller pore radius of 0.012 cm. This modification is naturally to be expected, since the breakdown of large crumbs of fairly uniform size produces smaller crumbs in a greater range of sizes, which causes a general reduction of pore size and a reduction of the prominence of any one group of pores of a particular size. After two cycles of wetting and drying there is still a perceptible group of pores, the peak now occurring at the further reduced pore radius of 0.007 cm. The mechanical analysis of this soil is given in table 1.

The curves of figures 5 and 5a show a similar series of moisture characteristics and differentials thereof for a sample of Oxford clay, of poor lime status and known to have an acid reaction. The group of large pores is not so prominent

as in the Gault clay, even in curve 1 for the crumbs in the first stage. This soil is apparently not sufficiently stable to withstand completely the first soaking which is a preliminary to the taking of the moisture characteristic. The peak of the pore size distribution occurs at a pore radius of 0.019 cm., which, by comparison with the corresponding value of 0.023 cm. for Gault clay, is additional evidence in support of this conclusion. One cycle of wetting and drying is as effective in disintegrating the crumbs as are two cycles in the case of Gault, the much reduced peak in the pore size distribution occurring at a pore radius of 0.008 cm. as compared with 0.007 cm. for Gault after two

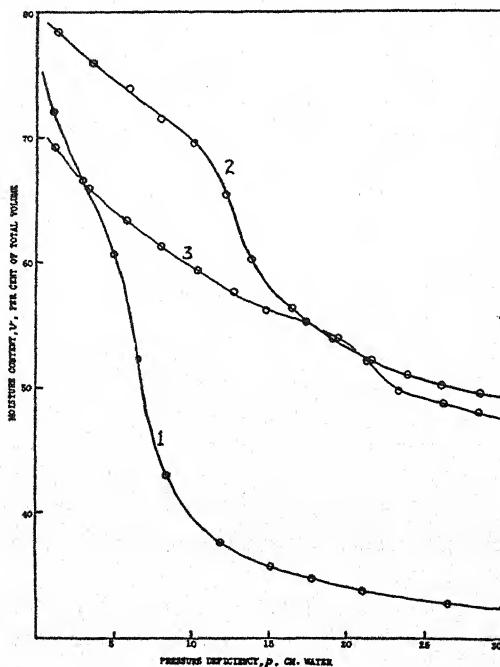


FIG. 4. MOISTURE CHARACTERISTICS OF GAULT CLAY CRUMB FRACTION, CRUMB SIZE INITIALLY BETWEEN 1 AND 2 MM.

Curve 1, immediately after separation by sieves.

Curve 2, after one cycle of wetting and drying.

Curve 3, after two cycles of wetting and drying.

cycles. After two cycles of weathering of the Oxford clay, one is unable to assert with confidence that there is any indication of a well-defined group of intercrumb pores. The mechanical analysis of this clay is also given in table 1.

The lack of stability of a third soil, Amphill clay, is shown by the series of moisture characteristics of figures 6 and 6a. The mechanical analysis given in table 1 would not lead one to expect this result, since the clay content is high and there is sufficient free calcium carbonate to prevent acidity. Nevertheless, it is seen that one cycle of "weathering" is sufficient to destroy any recognizable trace of a distinct intercrumb pore group.

It is impossible to discuss these results in terms of comparison between what is observed and what is expected, since it cannot be said that we have anything approaching a satisfactory understanding of the physics of soil binding, crumb

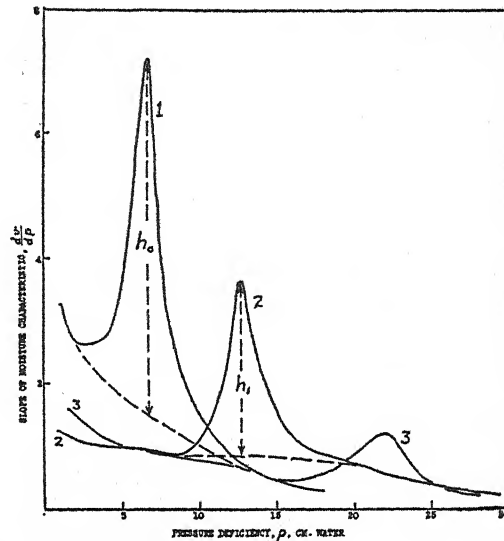


FIG. 4a. DIFFERENTIAL CURVES DERIVED FROM FIGURE 4, SHOWING PORE SIZE DISTRIBUTION

Curve 1, immediately after separation of crumb fraction, the marked peak at a pressure deficiency of 6.7 cm. water indicates particular abundance of pores of radius grouped about 0.023 cm.

Curve 2, after one cycle of wetting, the less marked peak at a pressure deficiency of 12.7 cm. water shows that the group of pores is still well defined, but the pore sizes are reduced, being grouped about 0.012 cm.

Curve 3, after two cycles of wetting and drying, the small peak at a pressure deficiency of 22.0 cm. water shows that a group of pores is still just discernible, the pore sizes now being grouped about the radius value of 0.007 cm.

Stability of Gault clay, $h_1/h_0 = 0.49$.

TABLE 1

Mechanical analyses of clay soils of different degrees of stability

SOIL	COURSE SAND	FINE SAND	SILT	CLAY	CaCO ₃
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Gault clay.....	3.9	7.4	17.7	61.1	7.9
Oxford clay.....	19.5	31.9	19.4	21.8
Amphill clay.....	1.0	3.4	20.5	71.0	0.14

formation, and soil stability in nature. Artificially produced crumbs, upon which a certain amount of work has been carried out in respect of binding forces and water stability, have very little in common with soil crumbs as found in the field. It is possible, however, in the light of the foregoing results,

to suggest a definition of soil stability sufficiently precise to enable one to assign a numerical value, in respect of this property, to any given soil. Con-

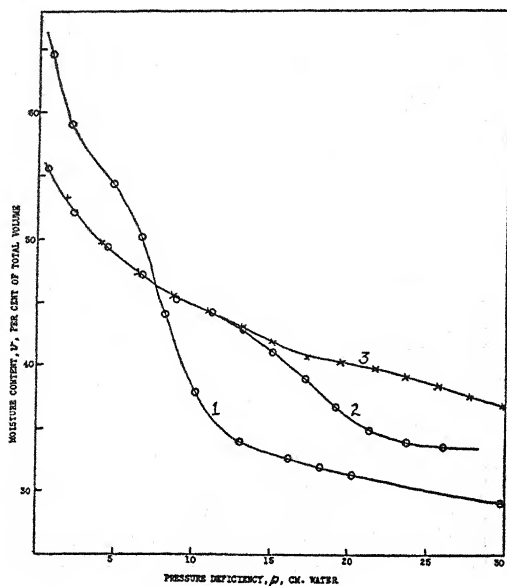


FIG. 5. MOISTURE CHARACTERISTICS OF OXFORD CLAY CRUMB FRACTION, CRUMB SIZE INITIALLY BETWEEN 1 AND 2 MM.

Curve 1, immediately after separation by sieves.

Curve 2, after one cycle of wetting and drying.

Curve 3, after two cycles of wetting and drying.

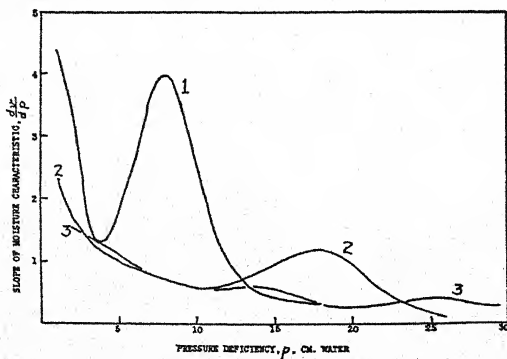


FIG. 5a. DIFFERENTIAL CURVES DERIVED FROM FIGURE 5, SHOWING PORE SIZE DISTRIBUTION

By comparison with figure 4a, it is seen that the stability is much less than that of Gault clay. The marked pore group at a pore radius of 0.019 cm. in curve 1 is much modified in curve 2 and is absent in curve 3.

Stability of Oxford clay = 0.28.

sider, for example, the curves of figure 4a. We can measure the height h of each peak above the general level of the curve. If h_0 is this height for the

first curve, and h_1 the height after one cycle of wetting and drying, then the ratio h_1/h_0 has the value unity for a perfectly stable soil, since the cycle has no effect on it, and zero for a completely unstable soil, since one cycle of

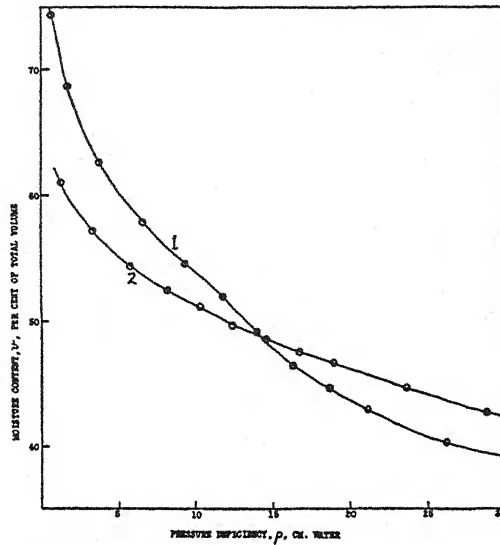


FIG. 6. MOISTURE CHARACTERISTICS OF AMPHTHILL CLAY CRUMB FRACTION, CRUMB SIZE INITIALLY BETWEEN 1 AND 2 MM.

Curve 1, immediately after separation by sieves.
Curve 2, after one cycle of wetting and drying.

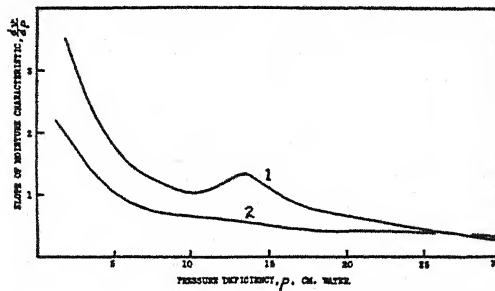


FIG. 6a. DIFFERENTIAL CURVES DERIVED FROM FIGURE 6, SHOWING PORE SIZE DISTRIBUTION

The small peak in curve 1 at a pressure deficiency of 13.6 cm. water indicates that there is only a slightly marked pore group at radius 0.011 cm.; this group has quite disappeared in curve 2. The soil is not sufficiently stable to withstand the first wetting and is extensively disintegrated by one cycle only of wetting and drying.

Stability of Amphthill clay = 0.

"weathering" of such soil eliminates the intercrumb pore space. We may therefore tentatively define this ratio as the numerical value of the soil stability. Thus for Gault clay, the stability is 0.49, for Oxford clay 0.28, and for Amphthill clay 0. Such a definition must be tentative, since further experience of many

different soils may show this stability scale to be too crowded at one end, probably the low stability end. The definition is, of course, based on an arbitrary method, and will not be of much value unless agreement can be reached on a standardized technic covering the crumb fraction selected and the process of wetting and drying. It does, however, open the way to further studies of the physical basis of crumb formation by permitting of a fairly precise comparison of soil stabilities.

GENERAL DISCUSSION OF MOISTURE CHARACTERISTICS

All the ascertainable facts about the relationship between soil moisture content and pressure deficiency are shown at a glance by the moisture characteristic. If well-defined groups of pore water, held by forces of different magnitude, exist in the soil, their presence will be indicated by points of inflection in the characteristic or by peaks in the differential curve; conversely, if such points of inflection or peaks are absent from the curves, then well-defined water masses are absent, and we are not justified in attempting to make a division of the soil water into classes on a physical basis. Thus a strict regard for the facts as revealed by the moisture characteristic imposes a discipline upon us. It is a fact that most soil moisture characteristics give no grounds for a physical system of soil water classification; we cannot choose a point on a normal characteristic and say with justification that the water to one side of this point is of one kind and that to the other side is of another kind. The conventional division of soil water into gravitational, capillary, and hygroscopic water, and so on, has no validity. The use of terms implying the possibility of such a division may be convenient and not dangerous when limited to general description, but any attempt at precise definition on any but an arbitrary basis is foredoomed to failure. To take an example, the definition of "moisture-holding capacity" as "that moisture content which the soil can retain against gravity when freely draining" is, as a soil constant, meaningless, since the value so defined can vary from saturation to air-dry moisture content, depending on the height of the soil above the free water level with which it is in equilibrium and to which any surplus water drains. Attention has been drawn in the past to the lack of scientific foundation to soil water classification (5, p. 211) but unfortunately this lack is still not generally admitted. As recently as 1939, for example, Zunker presented a formidable list of definitions of various classes of soil water (12). The general weakness of his case is illustrated by the definition of adsorbed water, or water hygroscopicity, as "Wasser, das an der Kornoberfläche durch den Oberflächendruck der Bodenteilchen angelagert und verdichtet ist," while the quantity of water so held is defined purely arbitrarily as "the quantity of water taken up by 100 gm. of dry soil over 10% sulphuric acid at room temperature of about 18°C." As an example of the legitimate drawing of a distinction between different masses of bound water, we may refer to the curves published by Kelley, Jenny, and Brown (6) showing the course of dehydration of various minerals.

If we are justified in interpreting all soil moisture characteristics as indicators of pore size distribution, it is at once apparent that there is no possibility of developing mathematical formulas on physical grounds, purporting to represent these curves. Expressions such as those of Vageler and Alten (1, 11) and of Gardner (3), insofar as they fit the facts, may perhaps be best regarded as empirical formulas representing common types of pore size distribution. A study of published moisture characteristics shows that the formulas mentioned are too simple to have general validity. Vageler and Alten write

$$p = 50(H_v/W)^3 \quad (B)$$

and

$$p = 50(H_v/W)^2 \quad (C)$$

in different papers, p being the pressure deficiency, H_v the hygroscopic capacity, and W the water content. In each formula the whole shape of the curve depends on only one soil parameter, the hygroscopic capacity. This one parameter therefore determines the whole curve, and if two moisture characteristics have one point in common, they must coincide everywhere. Gardner writes

$$p = A + B/W^3 \quad (D)$$

This has two arbitrary constants, A and B characteristic of the soil, and therefore permits two soil moisture characteristics to have one, and only one, point in common without coinciding everywhere. Schofield has, however, published a number of moisture characteristics (10), drawn from data given by Middleton (8), containing pairs (Clarksville colloid-Norfolk colloid and Clarksville colloid-Cecil colloid) which cross each other once and tend to do so again outside the range of the experiment. The pair of moisture characteristics for Sharkey colloid and Norfolk colloid, though not actually behaving in this way, show a sufficient tendency to do so to be in conflict with equations (B) to (D). It is possible that in some cases (e.g., for colloidal materials) the shape of the moisture characteristic may be determined by variation of osmotic pressure with moisture content, the osmotic pressure being due to dissociated cations. A rigid mathematical discussion might in this case be justified but would not be simple. As we have seen, simplicity in the final expression for pressure deficiency would be at variance with the facts.

SUMMARY

The name "soil moisture characteristic curve" is proposed for curves obtained by plotting soil moisture content against the suction pressure, or any convenient function of the latter, with which the soil water is held. The use of soil moisture characteristics in soil studies is illustrated by application to the study of tilth in the field and of soil stability. A definition of soil stability is proposed, enabling this property to be expressed by a numerical value. Finally, the classification of so-called types of soil water and mathematical expressions for soil water pressure deficiency are discussed in the light of known types of moisture characteristics.

REFERENCES

- (1) ALTEN, F. 1938 Zur Beurteilung des pflanzenaufnehmbaren Wassers aus dem Boden mit Hilfe des pF-Wertes. *Trans. First Conn. Internatl. Soc. Soil Sci.* A: 46-50.
- (2) FOSTER, A. G. 1932 The sorption of condensable vapours by porous solids. *Trans. Faraday Soc.* 28: 645-657.
- (3) GARDNER, W. 1919 The movement of water in soil by capillarity. *Soil Sci.* 7: 313-317.
- (4) HAINES, W. B. 1930 Studies in the physical properties of soil. *Jour. Agr. Sci.* 20: 97-116.
- (5) KEEN, B. A. 1931 The Physical Properties of the Soil. Longmans Green.
- (6) KELLEY, W. P., JENNY, H., AND BROWN, S. M. 1935 Hydration of minerals and soil colloids in relation to crystal structure. *Trans. Third Internatl. Cong. Soil Sci.* 3: 84-87.
- (7) LEAMER, R. W., AND LUTZ, J. F. 1940 Determination of pore size distribution in soils. *Soil Sci.* 49: 347-361.
- (8) MIDDLETON, H. E. 1927 The adsorption of water vapour by soils and soil colloids. *Proc. First Internatl. Cong. Soil Sci.* 1: 446-455.
- (9) NICHOLSON, H. H. 1935 The mode of action of mole drains. *Farm and Machine* 2: 61-67.
- (10) SCHOFIELD, R. K. 1938 Pore size distribution as revealed by the dependence of suction on moisture content. *Trans. First Conn. Internatl. Soc. Soil Sci.* A: 38-45.
- (11) VAGELER, P., AND ALTEN, F. 1931, 1932 Böden des Nil und Gash. *Ztschr. Pflanzen-ernähr., Düngung, u. Bodenk.* (A) 21: 47-57, 323-346; 22: 21-51, 191-267 (1931); 23: 149-207, 208-339; 24: 50-92, 179-242 (1932).
- (12) ZUNKER, F. 1938 Vorschläge für Begriffsbildungen in der Boden und Grundwasser-kunde. *Kulturtechniker* 41: 328-334. [Abs. in *Soil Res.* 6: 202-216 (1939).]

RELATIONSHIP BETWEEN ORGANIC MATTER CONTENT AND MOISTURE CONSTANTS OF SOILS¹

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Many line fences in Michigan were established at the time the land was cleared, or shortly afterward. Along these fence rows a continuous sod of bluegrass has existed, while a few feet away, in the adjoining fields, the customary rotations, with the usual tillage and other soil management practices, have been followed. Preliminary studies having shown that the soil under the sod was appreciably higher in organic matter than the soil in the adjoining fields, a more complete comparative study was undertaken.

METHODS

Samples were collected by means of a spade, from the surface 6 inches of soil, in the fall of 1936. These samples were air-dried, screened to remove gravel, and then thoroughly mixed. They include 3 loamy sands, 9 sandy loams, 8 loams, 2 silt loams, 1 clay loam, 1 loamy fine sand, and 1 fine sandy loam.

Hygroscopic moisture was calculated in the conventional manner, after drying at 110°C. Moisture-equivalent and wilting-coefficient determinations were made according to the methods of Bouyoucos.³ The percentages of available moisture were obtained by subtracting the wilting coefficients from the moisture equivalents.

Organic-matter content was estimated by calculation from the carbon content, as determined by means of the combustion chain. Data from direct ignition are included for comparison, but in the studies of the relationship between organic-matter content and moisture constants, only the values derived from the carbon determinations are used.

CONCLUSIONS

In all but one case the organic matter content of the sample collected from under sod was higher than that of the sample from the adjoining field.

¹ Contribution from the Soils Section of Michigan Agricultural Experiment Station. Authorized by the director for publication as journal article 451 n.s.

² Former graduate student in soils and graduate assistant in farm crops, respectively.

³ Bouyoucos, G. J. A comparison between the suction method and the centrifuge method for determining moisture equivalent of soils. *Soil Sci.* 40: 165-171, 1935; A mechanical device for determining the permanent wilting point of soils by means of the cohesion method. *Soil Sci.* 46: 331-335, 1938.

TABLE 1
Comparison of the moisture relations of pairs of soil samples

SAM- PLES*	SOIL CLASSES	HYGRO- SCOPIC MOISTURE	MOISTURE AT WILTING POINT	MOISTURE EQUIVA- LENT	AVAIL- ABLE MOISTURE	ORGANIC MATTER†	LOSS ON IGNITION
		<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
1a	Loamy sand	0.89	5.32	10.21	4.89	1.00	1.06
1b		1.76	7.83	19.34	11.51	3.17	3.09
2a		1.27	7.81	15.26	7.45	1.10	1.37
2b		0.79	8.09	19.12	11.03	1.91	2.04
3a		0.53	5.86	16.36	10.50	2.45	1.59
3b		0.92	5.68	11.73	6.05	3.82	1.77
4a	Loamy fine sand	0.92	10.03	21.58	11.55	2.40	2.42
4b		1.29	12.28	29.20	16.92	4.50	3.98
5a		0.33	5.68	13.29	7.61	2.15	1.63
5b		0.54	7.31	19.09	11.78	1.81	2.12
6a		0.99	13.73	23.42	9.69	3.88	3.47
6b		0.69	13.91	28.50	14.59	5.68	3.75
7a		0.91	8.11	19.08	10.97	1.86	2.50
7b		1.55	8.08	21.77	13.69	3.45	3.57
8a		0.52	5.37	11.98	6.61	1.26	2.18
8b		0.95	7.19	17.62	10.43	1.53	2.58
9a	Sandy loam	0.92	6.51	12.97	6.46	1.02	1.85
9b		0.59	7.75	16.11	8.36	3.61	2.42
10a		0.75	6.98	15.95	8.97	1.07	2.41
10b		0.76	8.71	21.93	13.22	1.30	2.04
11a		1.60	11.42	20.87	9.45	1.29	1.00
11b		2.09	11.05	20.60	9.55	1.77	1.37
12a		0.80	8.54	19.48	10.94	1.54	2.26
12b		1.10	8.71	21.29	12.58	1.95	2.95
13a		0.56	7.58	13.48	5.90	1.06	2.10
13b		0.98	8.07	18.61	10.54	1.76	3.38
14a	Fine sandy loam	0.80	8.54	19.48	10.94	1.54	2.26
14b		1.10	8.71	21.29	12.58	1.95	2.95

* *a* samples collected from cultivated field, and *b* samples from under sod along the adjoining fence row.

† Total carbon $\times 1.724$.

TABLE 1—*Concluded*

SAM- PLES*	SOIL CLASSES	HYGRO- SCOPIC MOISTURE	MOISTURE AT WILTING POINT	MOISTURE EQUIVA- LENT	AVAIL- ABLE MOISTURE	ORGANIC MATTER†	LOSS ON IGNITION
		<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
15a	Loam	1.39	9.09	19.94	10.85	2.93	3.98
15b		1.69	9.84	28.06	18.22	4.35	3.48
16a		3.07	11.06	25.15	14.09	4.37	4.40
16b		1.42	11.43	31.49	20.06	4.98	5.19
17a		1.17	8.56	21.96	13.40	1.41	3.31
17b		1.17	7.55	26.61	19.06	2.96	4.09
18a		1.25	8.54	22.41	13.87	1.24	3.21
18b		1.38	9.21	27.89	18.68	3.07	3.64
19a		1.22	8.88	21.86	12.98	2.71	3.51
19b		1.34	9.55	22.57	13.02	2.80	3.98
20a		0.82	11.26	22.36	11.10	2.83	2.28
20b		1.02	13.00	37.65	24.65	5.68	3.27
21a		2.40	13.18	30.52	17.34	1.29	3.53
21b		2.68	14.43	41.27	26.84	6.15	4.01
22a		2.92	13.40	31.19	17.79	3.89	3.86
22b		3.44	14.84	44.40	29.56	7.87	3.79
23a	Silt loam	1.43	11.60	23.70	12.10	3.69	2.79
23b		3.34	10.19	24.73	14.54	4.80	3.28
24a		0.90	8.41	18.80	10.39	1.93	2.59
24b		1.05	8.23	21.95	13.72	2.20	2.92
25a	Clay loam	3.99	11.78	27.31	15.53	2.36	2.98
25b		4.08	11.18	30.46	19.28	3.10	3.76
Average, <i>a</i> samples		1.29	9.09	19.94	10.85	2.09	2.58
Average, <i>b</i> samples		1.51	9.71	24.93	15.22	3.45	3.18

With two exceptions, the soil under sod had a higher moisture equivalent than the corresponding soil in the field.

In all but five samples, the hygroscopic moisture content of samples from fence rows was higher than that of corresponding samples from fields. In seven cases, the wilting coefficient of the field soil was higher than that of the corresponding soil under sod.

Available moisture content was found to be greater in the soil under sod, with one exception, than in the corresponding soil from the field.

A study of the data by means of Fisher's method⁴ showed a direct correlation between organic-matter content of the soils and their available moisture, with a correlation coefficient of 7.28.

⁴ Fisher, R. A. 1930 Statistical Methods for Research Workers, ed. 3. Oliver & Boyd, Edinburgh.

RAPID MICRODETERMINATION OF BORON BY MEANS OF QUINALIZARIN AND A PHOTOELECTRIC COLORIMETER

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The increased use of boron as a fertilizer and the recognition of an increasing number of physiological disorders in plants as boron deficiencies have resulted in the development recently of a number of methods for the determination of boron in the small amounts found in soils and plants (1-12). The electro-metric titration method using mannite has been applied successfully to soils and plant material of the arid or semiarid regions of the West where the concentration of boron is relatively high (10, 11). In the more humid regions, where smaller quantities of boron occur, spectrographic methods (3, 4, 6) and the quinalizarin method (1, 7) have been used. Because of the expense involved and the training necessary for the use of the spectrograph, the quinalizarin colorimetric method seems adapted for use in a greater number of the laboratories. Of the several factors involved in the quinalizarin determination which have received little attention, the adaptability of the method for use with a photoelectric colorimeter and the effects on the reaction of temperature, concentration of acid, and time of standing appear worthy of additional consideration. This study concerns chiefly the adaptation of the quinalizarin method of determining boron for use in a photoelectric colorimeter. The other factors mentioned will necessarily be considered.

The addition of a boron compound to some of the hydroxyanthraquinones in concentrated sulfuric acid will cause a color change which may be used to identify the quinones. Conversely, the addition of these quinones to a solution of boric acid in concentrated sulfuric acid may be used as a qualitative and quantitative test for boron. Of the various quinones that have been used for the determination of boron, quinalizarin (1,2,5,8-tetrahydroxyanthraquinone) has been found to be most suitable. The color changes from violet to blue with increasing amounts of boron. Oxidizing substances such as nitrates, dichromates, and permanganates will destroy the color of the quinalizarin and thus render the test useless. The sensitivity of the quinalizarin reaction is greatly affected by the concentration of sulfuric acid used. A concentration of 92 to 94 per cent by weight in the final solution appears to be most sensitive. In order to obtain this concentration in the final solution, it

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is necessary to use acid stronger than the ordinary 95.5 to 96.5 per cent by weight in carrying out the test. Tables 1 and 2 give the sensitivity of the reaction in two concentrations of sulfuric acid.

TABLE 1

Boron color standards with 98.5 per cent sulfuric acid (about 93 per cent in final solution)

NUMBER	AMOUNT AND STRENGTH OF BORON SOLUTION		BORON	OBSERVED COLOR
	cc.	p.p.m.	mgm.	
1	0		0	Violet
2	1	0.1	.0001	Violet
3	1	0.2	.0002	Violet tinged with blue
4	1	0.4	.0004	Violet gradually changing to blue
5	1	0.6	.0006	
6	1	0.8	.0008	
7	1	1.0	.0010	
8	1	2.0	.0020	
9	1	3.0	.0030	Blue
10	1	4.0	.0040	

TABLE 2

Boron color standards with 96 per cent sulfuric acid (about 91 per cent in final solution)

NUMBER	AMOUNT AND STRENGTH OF BORON SOLUTION		BORON	DISTILLED WATER	OBSERVED COLOR
	cc.	p.p.m.	mgm.	cc.	
1	0		0	1.0	Violet
2	0.5	1	.0005	0.5	Violet
3	1.0	1	.001	0	Violet tinged with blue
4	0.2	10	.002	0.8	Violet gradually changing to blue
5	0.3	10	.003	0.7	
6	0.4	10	.004	0.6	
7	0.5	10	.005	0.5	
8	0.6	10	.006	0.4	
9	0.7	10	.007	0.3	Blue
10	0.8	10	.008	0.2	
11	0.9	10	.009	0.1	
12	1.0	10	.010	0	Blue

VISUAL COLORIMETRIC METHOD

In carrying out the method as proposed by Smith (9), 1 cc. of the unknown or standard is pipetted into a convenient-sized glass tube, into which 9 cc. of 98.5 per cent sulfuric acid is run from a burette. The contents are stirred and cooled, and 0.5 cc. of a quinalizarin solution containing 0.010 gm. in 100 cc. of approximately 93 per cent sulfuric acid is added. After standing 15 minutes, the unknowns are compared with standards prepared in exactly the same manner. The final concentration of the sulfuric acid in this mixture is about 93 per cent by weight. One of the difficulties involved in this method is the

use of 98.5 per cent by weight sulfuric acid. This may be prepared by standardizing and mixing fuming sulfuric acid with ordinary 95.5 to 96.5 per cent sulfuric acid. As shown in table 2, when ordinary sulfuric acid is used, it is impossible to detect less than 0.002 mgm. of boron in 1 cc. volume, but with the 98.5 per cent acid (table 1), 0.0002 mgm. of boron in 1 cc. volume can be detected. By using the ordinary acid, the range is increased, and as much as 0.010 mgm. boron can be determined quantitatively; but when the more concentrated acid is used, amounts above 0.004 mgm. cannot be separated. In soil and plant analysis, the more concentrated acid is used. Aliquoting to reduce the sample is seldom necessary.

Since the volume of water and acid used in making up the standards is critical, all glassware measuring devices must be calibrated to deliver exactly the correct amount of solution.

Standards made up as described may be used for several days with reliable results, if they are tightly stoppered. No permanent color standards have yet been devised. The use of a photoelectric colorimeter saves considerable time in making up standards, for after a curve for a series of standards has once been established, checking at monthly intervals is sufficient to assure reliable results.

PHOTOELECTRIC COLORIMETRIC METHOD

Apparatus

The instrument used in this work is a Cenco-Sheard-Sanford photometer which employs filters and is calibrated in per cent light transmission. In the quinalizarin determination, its use is somewhat complicated because of the rather deep violet color of the blank. This blank color decreases the intensity of the light passing through the absorption cell to such an extent that it is impossible to get a very wide spread on the galvanometer scale. In the desired range of boron concentration, however, a spread of 15 to 20 divisions was obtained, and this was considered reasonably good. Since in the quinalizarin boron determination, the intensity of the blue color is measured, an orange filter is used.

Absorption cells

In preliminary work, fused pyrex absorption cells of 1 cm. thickness and 10 cc. capacity were used, but better sensitivity was obtained by using absorption cells of 5 cm. thickness and 50 cc. capacity. Deflections on the galvanometer scale of only 1.2 and 1.9 divisions with 0.0005 and 0.001 mgm. boron were obtained with the 1-cm. cells, whereas the 5-cm. cells showed deflections of 17.4 and 21.4 divisions, respectively, for the same amounts of boron. An interchangeable carriage may be obtained for the use of the larger absorption cells in the photometer.

Boron-free glassware should be used for all reagents. No appreciable blanks resulted from the use of pyrex absorption cells. Where solutions stood for

some time, it was necessary to use Kavalier boron-free glassware, aluminum, or copper for the containing vessels. In determinations involving small amounts of boron (about 0.02 mgm.), both pyrex and lime-glass gave blanks that were extremely high when hot solutions remained in them for 12 hours. Under these conditions, the blanks obtained by using different containers were as follows: pyrex, 0.03 mgm. boron; lime-glass, 0.005; Kavalier boron-free glass, 0.0005; and aluminum, 0.0006.

Reagents

Boron standards.

A. Standard containing 0.5 mgm. boron per cubic centimeter. Dissolve 2.8578 gm. of dried, reagent quality boric acid crystals in distilled water and make up to 1 liter.

B. Standard containing 0.01 mgm. boron per cubic centimeter. Dilute 20 cc. of standard A to 1000 cc. with distilled water.

C. Standard containing 0.001 mgm. boron per cubic centimeter. Dilute 100 cc. of standard B to 1000 cc. with distilled water.

Quinalizarin solution. Dissolve 0.20 mgm. dried quinalizarin in 98.5 per cent by weight sulfuric acid and make up to 1 liter with the same strength acid.

Sulfuric acid solutions.

A. 98.5 per cent by weight. This can be prepared by titrating fuming sulfuric and ordinary concentrated sulfuric acid with 1.0 *N* alkali and mixing appropriate quantities of the two. This alkali standardization is not necessary for routine work. Once the titration values for the same lot of fuming and the same lot of ordinary sulfuric acid are determined, it is not necessary to repeat the titration until a new lot of acid is obtained. The approximate appropriate mixture of the two acids, as determined by the single titration, may be standardized accurately by using it in making a photometer curve with standard boron solutions. In this work 500 gm. of fuming sulfuric acid mixed with each 2 liters of ordinary concentrated sulfuric acid was found to give the desired concentration. For convenience, this acid is made up in 5-gallon quantities and stored in smaller quantities in tightly stoppered bottles. For dispensing the acid, 5-gallon bottles equipped with a closed burette system and several bubbling bottles containing the same strength acid are used. Though this solution will keep well in such containers, it should be checked against boron standards at monthly intervals to see whether dilution has occurred as a result of absorption of moisture from the air.

B. Approximately 20 *N*. Add 1000 cc. of ordinary concentrated sulfuric acid to 800 cc. distilled water.

Procedure

Five cubic centimeters of the unknown or boron standard containing up to 0.03 mgm. boron is placed in a 125-cc. Erlenmeyer flask, and 5 cc. of the 20 *N* sulfuric acid and 50 cc. of the 98.5 per cent acid are added from burettes. The flasks are stoppered, thoroughly stirred, and cooled in a water bath to 80°F. One cubic centimeter of the quinalizarin solution is added, and the mixture is again stirred and allowed to stand for $\frac{1}{2}$ hour in the 80° water bath before being placed in the absorption cells of the photometer.

Temperature effect

The temperature in the laboratory where this work was carried out varied from approximately 65° during the winter months to about 90° during the summer. The effect of this temperature variation on the quinalizarin reaction

TABLE 3
Effect of temperature on the quinalizarin reaction

STANDARD BORON	PHOTELOMETER READINGS AT			
	70°F.	80°F.	90°F.	100°F.
<i>mgm.</i>				
0	56.8	57.1	58.5	60.0
.0025	52.9	53.6	55.5	57.2
.0050	51.8	52.2	53.0	54.9
.0075	47.8	50.0	52.0	53.7
.0100	45.1	47.4	49.5	52.3
.0125	44.7	46.2	48.5	51.0
.0150	41.8	44.3	47.5	50.0
.0200	40.6	42.7	45.8	47.1
.0250	38.5	40.6	42.3	45.3
.0300	37.6	39.5	41.8	42.5

was studied by cooling or warming the solutions in a water bath before making the determinations. These results are shown in table 3. The rise in temperature from 65 to 90°F. produces a very noticeable drift upward in the photometer readings. These data indicate that the control of temperature in the quinalizarin determination of boron is desirable.

Reproducibility of results

Under the same conditions of temperature, different standards were made up and run at approximately 1-week intervals to determine the reproducibility of values obtained. Table 4 gives these results. The largest difference between any two of these readings is 1.0. In practice, duplicate determinations should be made and when a standard or unknown does not check within 0.5, the determination should be repeated. Figure 1 shows the calibration curve obtained by plotting the average values for the three sets of standards.

Concentration of sulfuric acid

The sensitivity of the quinalizarin reaction, as mentioned previously, is somewhat decreased when ordinary sulfuric acid is used instead of 98.5 per

TABLE 4

Reproducibility of photometer readings for three boron standards prepared at 1-week intervals at 80°F.

STANDARD BORON	PHOTELOMETER READINGS			
	A	B	C	Average
<i>mgm.</i>				
0	57.5	57.0	57.5	57.3
.0025	53.5	53.8	54.5	53.6
.0050	52.5	52.0	52.0	52.2
.0075	50.3	49.5	50.2	50.0
.0100	47.0	47.2	47.9	47.4
.0125	45.8	46.1	46.8	46.2
.0150	44.0	43.8	45.0	44.3
.0200	42.2	42.8	43.2	42.7
.0250	40.5	40.2	41.0	40.6
.0300	39.8	39.3	39.5	39.5

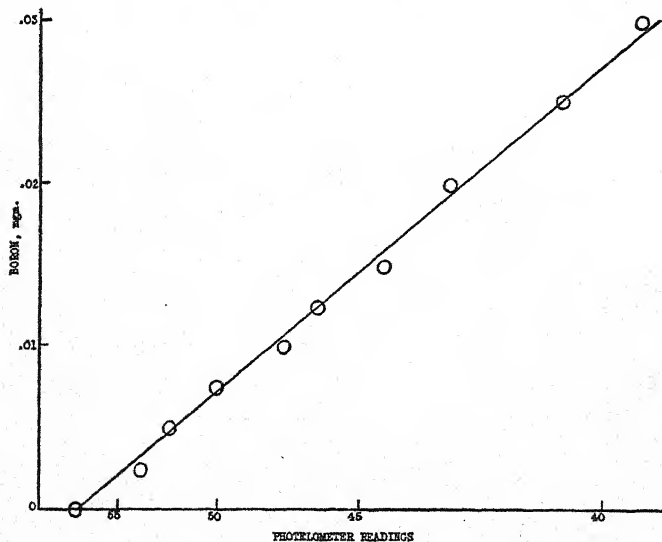


FIG. 1. CALIBRATION CURVE FOR BORON STANDARDS

cent acid. Table 5 gives the results obtained with the photometer using these two concentrations of acid with standards containing from 0 to 0.03 mgm. boron. The decrease in sensitivity with the less concentrated acid is considerable when less than .005 mgm. boron is present.

Time of standing

To determine whether one-half hour of standing was sufficient for maximum color development, photometer readings were obtained at $\frac{1}{2}$ hour and at 24 hours on the same set of standards. The data obtained are reported in table 6. The conclusion drawn from these data is that $\frac{1}{2}$ hour's standing is sufficient. Twenty-four hours of standing did not further intensify the color of the reaction.

TABLE 5

Effect of two concentrations of sulfuric acid on the sensitivity of the quinalizarin reaction

STANDARD BORON	PHOTOMETER READINGS	
	98.5 per cent H ₂ SO ₄ *	96.0 per cent H ₂ SO ₄ †
<i>mgm.</i>		
0	57.3	56.8
.005	52.2	55.8
.010	47.4	52.1
.015	44.3	49.6
.020	42.7	48.0
.030	39.5	44.0

* About 91 per cent in final concentration.

† About 89 per cent in final concentration.

TABLE 6

Effect of time of standing on the intensity of the color produced in the quinalizarin reaction

STANDARD BORON	PHOTOMETER READINGS	
	$\frac{1}{2}$ hour	24 hours
<i>mgm.</i>		
0	57.3	57.8
.0025	53.6	54.0
.0050	52.2	52.3
.0075	50.0	50.6
.0100	47.4	47.9
.0125	46.2	46.6
.0150	44.3	44.8
.0200	42.7	43.5
.0250	40.6	40.8
.0300	39.5	39.1

REFERENCES

- (1) BERGER, K. C., AND TRUOG, E. 1939 Boron determination in soils and plants. *Indus. and Engin. Chem., Analyt. Ed.* 11: 540-545.
- (2) COOK, R. L., AND MILLAR, C. E. 1938 A microtitration method for determining the readily soluble boron in soils. *Proc. Soil Sci. Soc. Amer.* 3: 146-152.
- (3) MCHARGUE, J. S. 1933 Report of less common metals in plants. Spectroscopic method for determination of boron in plant material. *Jour. Assoc. Off. Agr. Chem.* 16: 465-471.

- (4) MCHARGUE, J. S., AND CALFEE, R. K. 1932 Determination of boron, spectrographically. *Indus. and Engin. Chem., Analyt. Ed.* 4: 385-394.
- (5) NAFTEL, J. A. 1939 Colorimetric microdetermination of boron. *Indus. and Engin. Chem., Analyt. Ed.* 11: 407-409.
- (6) ROGERS, L. G., et al. 1939 Distribution of macro and micro elements in some soils of peninsular Florida. *Fla. Agr. Exp. Sta. Tech. Bul.* 341.
- (7) SCHARRER, K., AND GOTTSCHALL, R. 1935 Methods for the determination of boric acid in traces. *Ztschr. Pflanzenernähr., Düngung, u. Bodenk.* 39: 178.
- (8) SCOTT, W. W., AND WEBB, S. K. 1932 Determination of minute amounts of boron in soils. *Indus. and Engin. Chem., Analyt. Ed.* 4: 180-181.
- (9) SMITH, G. S. 1935 Determination of small amounts of boron by means of quinalizarin. *Analyst* 60: 735-739.
- (10) WILCOX, L. V. 1930 Determination of boron in natural waters and plant materials. *Indus. and Engin. Chem., Analyt. Ed.* 2: 358-361.
- (11) WILCOX, L. V. 1940 Determination of boron in plant materials. *Indus. and Engin. Chem., Analyt. Ed.* 12: 341-343.
- (12) WOODBRIDGE, C. G. 1937 Boron content of apple tissues as related to drought spot and corky core. *Sci. Agr.* 18: 41-48.

FACTORS AFFECTING AGGREGATION OF CECIL SOILS AND EFFECT OF AGGREGATION ON RUN-OFF AND EROSION¹

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It is recognized that different soils under the same environmental conditions vary in erodibility because of inherent differences in soil properties. These differences may be physical, chemical, or biological and may be either natural or artificial. The kind of macroflora and microflora, the kind of fertilizers used, and the cultural practices, such as plowing and disking, undoubtedly produce changes in the soil properties which influence erosion. It has been observed by Lutz (11) and determined by Yoder (23) that one of the factors influencing erosion is the degree to which the finer fractions (silt and clay) form water-stable aggregates. In order to develop and maintain a good state of aggregation in soils, it is necessary to know something about the factors causing aggregation.

These investigations were undertaken in an attempt to learn what these factors are and to ascertain more definitely the relationship that may exist between aggregation and soil erosion.

REVIEW OF LITERATURE

It has been shown (11) that the A and B horizons of a relatively nonerodible soil were highly granulated into large, stable, and porous aggregates; this was in contrast to the lower content of small, compact granules and the dispersed condition of the Iredell soils, which are erodible. The Cecil soils studied in these investigations occur, as do the Iredell and Davidson, in the Piedmont section of North Carolina and adjacent states. Chemically, the Cecil and Davidson are very similar and are more lateritic than the Iredell. The Cecil soils, however, are less aggregated than the Davidson (13). Peele's data (16) showed that untreated Cecil soils have less than 35 per cent of the silt plus clay aggregated, but Discker and Yoder (8) found an extremely high degree of aggregation in the Cecil.

Peele (17) found that lime had a dispersing effect on the aggregates and rendered the soil less permeable to water, thereby increasing run-off and susceptibility to erosion. The addition of organic matter had a beneficial effect on the soil, resulting in better aggregation and an increase in permeability. Peele (18) reported that soils having sandy loam surfaces may have a higher percentage of run-off and be more susceptible to erosion than soils with clay

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surfaces. Yoder (23) studied the physical nature of the erosion process on carefully controlled plots of Cecil clay on different slopes. His data showed, under all conditions of his work, that the larger aggregates eroded considerably less than the smaller ones.

Browning (7) reported that organic matter decreased the dispersion ratio and increased the number of larger sized aggregates when applied to soils of relatively poor structure containing only a small amount of cementing material necessary for the formation of stable aggregates. When the organic matter was added to soils containing appreciable quantities of either active inorganic or organic colloidal material, the dispersion or aggregation was not appreciably changed. He found that lime, in addition to organic matter in certain soils, materially reduced the percolation rate of the laboratory-packed samples.

Harper (9), discussing soil structure and moisture movement, pointed out that when relatively large quantities of organic matter are present in a soil, the physical condition is favorable for the absorption of water, regardless of soil granulation.

Baver (2) classified the aggregating constituents of soils into organic matter, divalent cations, and iron and aluminum. From a study of 77 soils throughout the United States, exclusive of lateritic soils, he concluded that organic matter is the major cause of stable soil granulation. Lutz (13) showed that free iron in the soil is an important factor in the granulation of lateritic soils.

Flocculation is perhaps the first step in granulation. The factors affecting flocculation should, therefore, throw considerable light on the problem of granulation. Baver (1) studied the effect of substituted cations on the physical properties of soil colloids and found that secondary particles of hydrogen clay were dispersed by substituted calcium ions. Lutz (12) found that hydrogen-clay membranes were more permeable to water than calcium-clay membranes. The hydrogen clays were less hydrated than the calcium clays. Bradfield (6) cited Paulding clay as having the poorest physical condition of any soil in Ohio in spite of the fact that it contains a fairly high amount of lime and organic matter. Myers (15) studied some physical properties of H- and Ca-saturated organic and inorganic colloids and found that the hydrogen systems had a greater degree of aggregation than corresponding calcium systems. These results are similar to those of Baver (3), who found that hydrogen soils formed by removing the calcium by acid leaching were as well aggregated as the original soils.

In discussing the reaction and buffering properties of humus, Waksman (22) says: "In combination with calcium or magnesium, humus is practically neutral or slightly alkaline, having a reaction of pH 7.0-7.5. In the absence of calcium and magnesium humus combines with aluminum hydroxide, iron hydroxide, or clay fractions, these being acid in reaction (pH 3.8). The first form of humus is saturated with bases and has little protective action for colloids; the acid form is unsaturated, liberates hydrogen when allowed to interact with neutral salts, and exerts a protective action upon colloids. One need not think of two forms of humus, but of two states of the same general type of humus."

Baver and Hall (4) and Myers (15) reported that dehydrated calcium-organic colloid was much more readily dispersed in water than dehydrated hydrogen-organic colloid. Baver and Hall (4), in a study of the influence of various cations on the flocculation of colloidal humus systems, found that the hydrogen ion is a more powerful flocculating agent than the barium or calcium ions.

EXPERIMENTAL METHODS

Soils from several plots of the U. S. Soil Conservation Experiment Station, Statesville, and from the Statesville Branch Station of the North Carolina Agricultural Experiment Station were used for these investigations. They will be referred to as the Conservation Station soil and the Fertility Station soil, respectively. A number of physicochemical properties of these soils were investigated, and the results are given in two parts: part I, the Conservation Station soil; and part II, the Fertility Station soil.

Part I—Conservation Station soil

Soil samples from the Conservation Experiment Station plots were selected in order that the physicochemical data could be interpreted with reference to run-off and erosion (table 3). The plots were established in 1930 on Cecil sandy clay loam with a 10 per cent slope. Various fertilizer and cropping treatments were used on the $\frac{1}{10}$ -acre plots (tables 1 and 2). Soil samples from plots 5 to 10 inclusive of the duplicate control series were taken in the fall of 1937 for these investigations. The samples were air-dried sufficiently to pass through a 2-mm. sieve and were stored in sealed jars. Moisture determinations were made, and all data are reported on the oven-dry basis.

TABLE 1
Cropping treatments of duplicate control plots of the Conservation Station

PLOT NUMBER	1931	1932	1933	1934	1935	1936	1937
5	S. corn F. wheat	S. wheat S. lesp.	S. lesp.	S. cotton	S. corn F. wheat	S. wheat S. lesp.	S. lesp.
6	S. wheat S. lesp.	S. lesp.	S. cotton	S. corn F. wheat	S. wheat S. lesp.	S. lesp.	S. cotton
7	S. lesp.	S. cotton	S. corn F. wheat	S. wheat S. lesp.	S. lesp.	S. cotton	S. corn F. wheat
8	S. cotton	S. corn F. wheat	S. wheat S. lesp.	S. lesp.	S. cotton	S. corn F. wheat	S. wheat S. lesp.
9	S. grass	grass	grass	grass	grass	grass	grass
10	S. cotton	S. cotton	S. cotton	S. cotton	S. cotton	S. cotton	S. cotton

S = Spring; F = Fall; Lesp. = Lespedeza.

The following determinations were made on the A and B horizons of these soils: aggregate analysis by a modification of the Kopecky elutriator method (5); organic carbon by the wet oxidation method of Tiurin (20); exchangeable hydrogen and total exchange capacity by the barium acetate method; the percentage of sesquioxides, obtained by leaching with 500 cc. of 0.075 *N* HCl (the soils were leached with .075 *N* HCl to remove the barium in the total exchange capacity determination); the pH electrometrically by using the glass electrode on 1:2 soil-water suspensions.

The data on the soil of the Conservation Station plots are shown in table 4. Unfortunately, no data were collected on the aggregation of the soil of these plots prior to the beginning of the treatments. All plots, however, were uniform with respect to previous treatments, and the differences that exist now are assumed to be due to the treatments. Aggregation was highest on plots

5 and 6, where a 4-year rotation, including lespedeza, was grown. Plots 7 and 8 had the same rotation, but this was started with a different crop in 1931, and, therefore, these plots had only three crops of lespedeza during the 7 years, whereas plots 5 and 6 had four (table 1). The slightly lower aggregation on plots 7 and 8 might also have been due to a longer time interval between turning lespedeza and sampling. The difference between plots 5 and 6 as compared with 7 and 8 is about 9 per cent in favor of plots 5 and 6. This indicates the value of a leguminous green manure crop in increasing the granulation of the Cecil soil. Erosion data show that plots 5 and 6 lost less soil during the 7-year period than plots 7 and 8. It should be observed,

TABLE 2
Fertilizer treatments of duplicate control plots of the Conservation Station

PLOT NUM- BER	1931	1932	1933	1934	1935	1936	1937	TOTAL LBS. PER ACRE		
								N	P ₂ O ₅	K ₂ O
5	400 lbs. 5-10-3	None	None	None	400 lbs. 4-10-4	400 lbs. 4-10-4	None	52	120	44
6	400 lbs. 2-10-4	None	None	None	None	None	600 lbs. 4-10-4	32	100	40
7	400 lbs. 2-10-4	None	None	None	None	600 lbs. 4-10-4	400 lbs. 4-10-4	48	140	56
8	600 lbs. 5-10-3	None	None	None	600 lbs. 4-10-4	400 lbs. 4-10-4	400 lbs. 4-10-4	86	200	74
9	400 lbs. 2-10-4	None	None	None	None	None	None	8	40	16
10	600 lbs. 5-10-3	None	None	None	600 lbs. 4-10-4	600 lbs. 4-10-4	600 lbs. 4-10-4	102	240	90

however, that plots 5 and 6 had only three row crops, whereas plots 7 and 8 had four. The greater degree of aggregation on plots 5 and 6 should result in less run-off and erosion even when all plots are in the same crop.

Plot 9 was in continuous grass during the 7-year period. A mixture was seeded at the beginning, but toward the end of the period orchard grass, which is shallow rooted, predominated. The data show that it was almost as effective as the 4-year rotation in aggregating the topsoil but, being shallow rooted, it had less effect on the subsurface soil. The 4-year rotation plots were cultivated in cotton and corn and prepared for wheat, whereas the continuous grass plot was not cultivated during the 7 years. It has been shown that cultivation destroys aggregates (10), and, therefore, the greater aggregation on the rotation plots appears even more significant.

TABLE 3

*Run-off and erosion data from the Statesville Soil Conservation Experiment Station, 7 years, 1931-1937, inclusive**

PLOT	RUN-OFF		EROSION					
	Inches per year	Per cent of rainfall	Lbs. soil per cu. ft. of run-off	Per cent Plot 4 = 100	Tons of soil per acre per year	Per cent Plot 4 = 100		
1	5.06	10.59	3.08	122	28	46		
2	3.64	7.61	1.77	70	12	19		
3	3.04	6.35	3.00	118	17	27		
4	14.20	29.69	2.53	100	62	100		
5	5.01	10.48	1.25	49	11	18		
6	4.11	8.61	1.73	68	11	18		
7	4.67	9.79	1.65	65	14	22		
8	4.36	9.11	1.65	65	13	21		
9	0.13	0.22	0.05	<1	<1	<1		
10	4.75	9.94	2.67	105	26	42		
11	4.42	9.24	2.93	116	28	45		
12	6.30	13.18	2.49	98	26	41		
	1931	1932	1933	1934	1935	1936	1937	Ave.
Rainfall, inches.....	44.98	55.28	34.70	49.34	43.58	60.00	47.52	47.80

Soil type, Cecil sandy clay loam.

Slope, 10 per cent.

* By personal communication from F. O. Bartel, then superintendent of the station. As mentioned in the context, only soils from plots 5 to 10 inclusive were used in these investigations.

TABLE 4

Laboratory data on the Conservation Station plots

PLOT NUMBER	PER CENT SILT + CLAY AGGREGATED	PER CENT ORGANIC CARBON	pH	PER CENT R_2O_3	TOTAL EXCHANGE CAPACITY M.E. PER 100 GM.	EXCHANGEABLE HYDROGEN M.E. PER 100 GM.
<i>A horizon</i>						
5	67.4	.701	5.45	.186	6.43	1.87
6	67.8	.624	5.50	.141	6.28	1.29
7	63.2	.642	5.33	.214	6.41	1.98
8	61.5	.679	5.31	.252	7.57	2.04
9	64.7	.789	5.58	.230	7.53	2.91
10	50.6	.682	5.54	.238	7.92	1.46
<i>B horizon</i>						
5	63.7	.402	5.41	.119	5.20	0.62
6	60.4	.262	5.13	.094	5.17	0.56
7	52.0	.277	5.24	.119	5.23	0.56
8	65.0	.284	5.22	.194	5.71	0.67
9	50.7	.259	5.31	.189	5.94	1.40
10	41.6	.271	5.38	.199	6.32	0.56

The continuous cotton plot, number 10, had the lowest percentage of aggregation, in both the topsoil and the subsurface soil. This clearly illustrates the detrimental effect on soil structure of continuous cultivation. The soil loss was greater on plot 10 than on any of the others studied. Undoubtedly, this was partly due to the lower degree of aggregation, because a year-by-year comparison of the erosion on plot 10 with that on the rotation plots when they were in cotton shows a greater loss on the continuous cotton plot.

The differences in aggregation are undoubtedly caused, in part at least, by the different crops and different amounts of cultivation. There are, however, several other factors that should be considered:

First, the aggregation decreased as the amount of P_2O_5 added increased on the cultivated plots (table 2). The P_2O_5 was applied as superphosphate, which means that calcium was applied in proportion to P_2O_5 . Peele (17) has asserted that calcium has a dispersing effect on Cecil clay, and it is possible that it has decreased the aggregation of the soil of these plots. The cation exchange capacity of the cultivated plots increased as the amount of fertilizer increased. Merkel (14) and Prince and Toth (19) have shown that the addition of phosphates to a soil increases its cation-exchange capacity. Toth (21), who studied the anion adsorption of the Cecil colloid, reported that the adsorption of silicates and phosphates reduced the free iron oxide content and resulted in an increased cation exchange capacity. It is possible that the phosphate reduced the amount of free iron oxide in these plots and thereby reduced aggregation, since it has been shown that aggregation of southern soils is correlated with the free iron oxide content (13). This idea is substantiated by the plant response to phosphate applications; the smaller amounts are tied up as insoluble iron phosphate, but the larger applications give a good response, indicating that all soluble iron has been precipitated.

Second, the different amount and kind of organic matter added by the different crops undoubtedly caused a difference in the microflora of the soil, which affected aggregation directly or possibly indirectly by modifying the soil humus. It has been observed that aggregates, as measured, may be held together by the organisms. No attempt has been made, however, to determine the number or kind of microorganisms present.

Part II—Fertility Station Soil

A series of fertilizer experimental plots was established on the Statesville Branch Station of the North Carolina Agricultural Experiment Station in 1910. The soil series, Cecil, was the same as that of the Conservation Station plots. A 4-year rotation of cotton, corn, wheat, and red clover was used, and different amounts and ratios of fertilizer were applied to the various plots. No clover was obtained during the first two rotations, 1910 to 1918; the plots were divided, therefore, and to the west end of each were applied 2 tons of lime per acre in 1919 and 1 ton per acre in each rotation thereafter, or a total of 6 tons per acre from 1918 to 1937.

No run-off and erosion data are available, but these plots were considered ideal for a study of the effects of the different fertilizer and liming treatments on aggregation for two reasons: first, aggregation has been shown to affect erosion, and it was felt that a study of these plots might provide data explaining the factors that cause aggregation; and second, noticeable differences in the physical condition of the soil indicated that lime and fertilizer had influenced the degree of aggregation either directly or by producing different amounts of organic matter. The six plots chosen for the investigation represent the extremes in physical condition and also three different fertilizer treatments. The fertilizer treatments are shown in table 5.

TABLE 5
Fertilizer treatments of Fertility Station plots

PLOT NUMBER		POUNDS PER ACRE			
		Limestone	N	P ₂ O ₅	K ₂ O
8 E	Cotton	None	15	42	15
	Corn		12	28	6
	Wheat		12	28	6
	Red clover		4	32	16
8 W	One ton of dolomitic limestone once in a rotation. Fertilizer treatment same as 8 E				
18 E	No lime No fertilizer				
18 W	One ton of dolomitic limestone once in a rotation. No fertilizer				
21 E	Cotton	None	30	84	30
	Corn		24	56	12
	Wheat		24	56	12
	Red clover		8	64	32
21 W	One ton of dolomitic limestone once in a rotation. Fertilizer same as 21 E				

The data in table 6 indicate that aggregation of the Cecil soil is affected by lime and by the amount and condition of the organic matter. It is evident that liming has decreased the aggregation on plots 18 and 21. This does not hold for plot 8, limed, on which the aggregation is 73.7 per cent as compared to 72.1 on plot 8, unlimed.

As shown in table 7, plot 8, unlimed, had a greater amount of exchangeable H than any other plot. This is an indication that the humus formed on it may have been of the "acid type." According to Waksman (22, p. 9), it takes many years of decomposition for "acid humus" to change to "mild humus." It may be thought that the increase in aggregation on plot 8, limed, was caused by the increase in organic matter (17 per cent) over that of plot 8,

unlimed. However, plot 21, limed, had 11 per cent more organic matter with 9.7 per cent less aggregation than plot 21, unlimed. For this reason it appears that the condition of the organic matter, rather than the amount, has the greater influence on aggregation. This is further substantiated by a comparison of plots 8 and 21, unlimed; both plots had the same degree of aggregation, but plot 21 had 27 per cent more organic matter and 1 m.e. more exchangeable calcium than plot 8. Apparently calcium-saturated humus is not so effective in aggregating the Cecil soil as is hydrogen- or sesquioxide-saturated humus.

In the A horizon, the unlimed part of plots 18 and 21 had more exchangeable hydrogen and more aggregation than the limed part. In the B horizon of

TABLE 6
Aggregation and other data on soil from Fertility Station plots

PLOT NUMBER	PER CENT SILT + CLAY AGGREGATED	PER CENT ORGANIC CARBON	pH	PER CENT R_2O_3
<i>A horizon</i>				
18 E	49.2	.392	5.43	.101
8 E	72.1	.652	5.25	.248
21 E	72.2	.826	5.65	.263
18 W	40.8	.394	7.15	.087
8 W	73.7	.765	6.98	.232
21 W	62.5	.918	6.78	.221
<i>B horizon</i>				
18 E	79.4	.218	5.04	.097
8 E	90.0	.252	5.16	.145
21 E	78.0	.381	5.47	.139
18 W	81.4	.308	6.43	.138
8 W	84.7	.265	6.31	.163
21 W	82.0	.474	5.92	.127

these plots, the limed part had more exchangeable hydrogen and a higher percentage of aggregation than the unlimed part. The B horizon of the unlimed part of plot 8 had a greater degree of aggregation and more exchangeable hydrogen than the limed part. These data all seem to indicate that good aggregation of Cecil soils is associated with a large amount of exchangeable hydrogen, and reducing the exchangeable hydrogen by liming or other means apparently destroys the aggregates, though if the soil has become very acid considerable time may be required.

Iron and aluminum are thought to play an important role in the aggregation of soils. Leaching the A horizons of both series with 0.05 *N* HCl gave more R_2O_3 from plots 8 and 21 than from plot 18. Plots 8 and 21 contained more organic matter than plot 18, and it is assumed that more of the iron and aluminum were combined as humates. Plots 8 and 21 also showed con-

siderably greater aggregation. In comparing individual plots of the limed and unlimed series it is seen that the A horizon of plots 18 and 21 gave less R_2O_3 on the limed than on the unlimed series, and the limed series was less aggregated. Waksman has pointed out that the R_2O_3 combines with humus in the absence of calcium and magnesium. These data strongly indicate that iron and aluminum humates are more influential in producing aggregation than are calcium and magnesium humates. In fact, it appears that humus saturated with calcium and magnesium is a poor aggregating agent. This is supported by observations of Bradfield (6) on Paulding clay.

TABLE 7
Base-exchange data on soil from Fertility Station plots

PLOT NUMBER	BARIUM ACETATE METHOD				AMMONIUM ACETATE METHOD			
	Total ex- change capacity	Exchange- able H	Fe	Al	Ca	Mg	K	Na
<i>A horizon</i>								
18 E	2.59	0.99	.030	.068	0.78	0.12	.17	.10
8 E	5.52	2.28	.058	.059	1.72	0.14	.33	.13
21 E	5.84	1.20	.023	.078	2.75	0.17	.33	.14
18 W	3.25	0.12	.012	.039	2.35	1.34	.05	.11
8 W	6.85	0.24	.024	.059	4.09	1.91	.23	.14
21 W	6.54	0.19	.016	.039	4.02	1.41	.18	.16
<i>B horizon</i>								
18 E	3.13	0.94	.024	.088	1.07	0.20	.20	.14
8 E	4.92	1.15	.036	.088	2.08	0.43	.22	.26
21 E	5.43	0.75	.026	.059	2.68	0.15	.27	.17
18 W	4.49	1.03	.012	.039	1.76	1.53	.13	.16
8 W	4.44	0.45	.016	.044	1.97	1.41	.14	.23
21 W	5.61	1.62	.016	.035	1.85	1.12	.09	.32

SUMMARY AND CONCLUSIONS

A study was made of the factors affecting the aggregation of Cecil soils. The relation between aggregation and erosion was investigated, and it was shown that better aggregation resulted in less soil erosion. Soils for these investigations were taken from the duplicate control plots of the Statesville Soil Conservation Experiment Station and from the Statesville Branch Station of the North Carolina Agricultural Experiment Station. The Conservation Station soil had been under experimentally controlled conditions for 7 years, and the Fertility Station soil for 28 years. A number of determinations, including percentage of silt plus clay aggregated, pH, exchangeable cations, total exchange capacity, and organic carbon, were made.

The Conservation Station soil showed that a crop rotation resulted in better aggregation with less erosion than continuous cotton cultivation. Also,

the inclusion of lespedeza in the rotation on two plots resulted in greater aggregation than that on a plot of a continuous sod of shallow-rooted grass. The addition of calcium through superphosphate resulted in decreased aggregation.

The Fertility Station soil showed that the addition of calcium through superphosphate or of calcium and magnesium through liming decreased aggregation, apparently by forming calcium or magnesium humates, which seem to be poorer granulating agents than iron, aluminum, or hydrogen humates. Hydrogen and sesquioxide humates increased aggregation regardless of the amount of organic matter, thus indicating that the condition of the humus is more important than the total amount in causing aggregation.

The results of these investigations on Cecil soils, which are lateritic, seem to warrant the following conclusions:

Good aggregation is beneficial in reducing run-off and erosion.

The condition of the organic matter is more important than the total amount in causing aggregation. Soils of the Cecil series are better aggregated when the humus is combined with hydrogen or sesquioxides than when combined with calcium or magnesium.

Liming decreased the aggregation of the soil of the Fertility Station plots in all but one case. This exception was explained as a residual effect of the original acid humus.

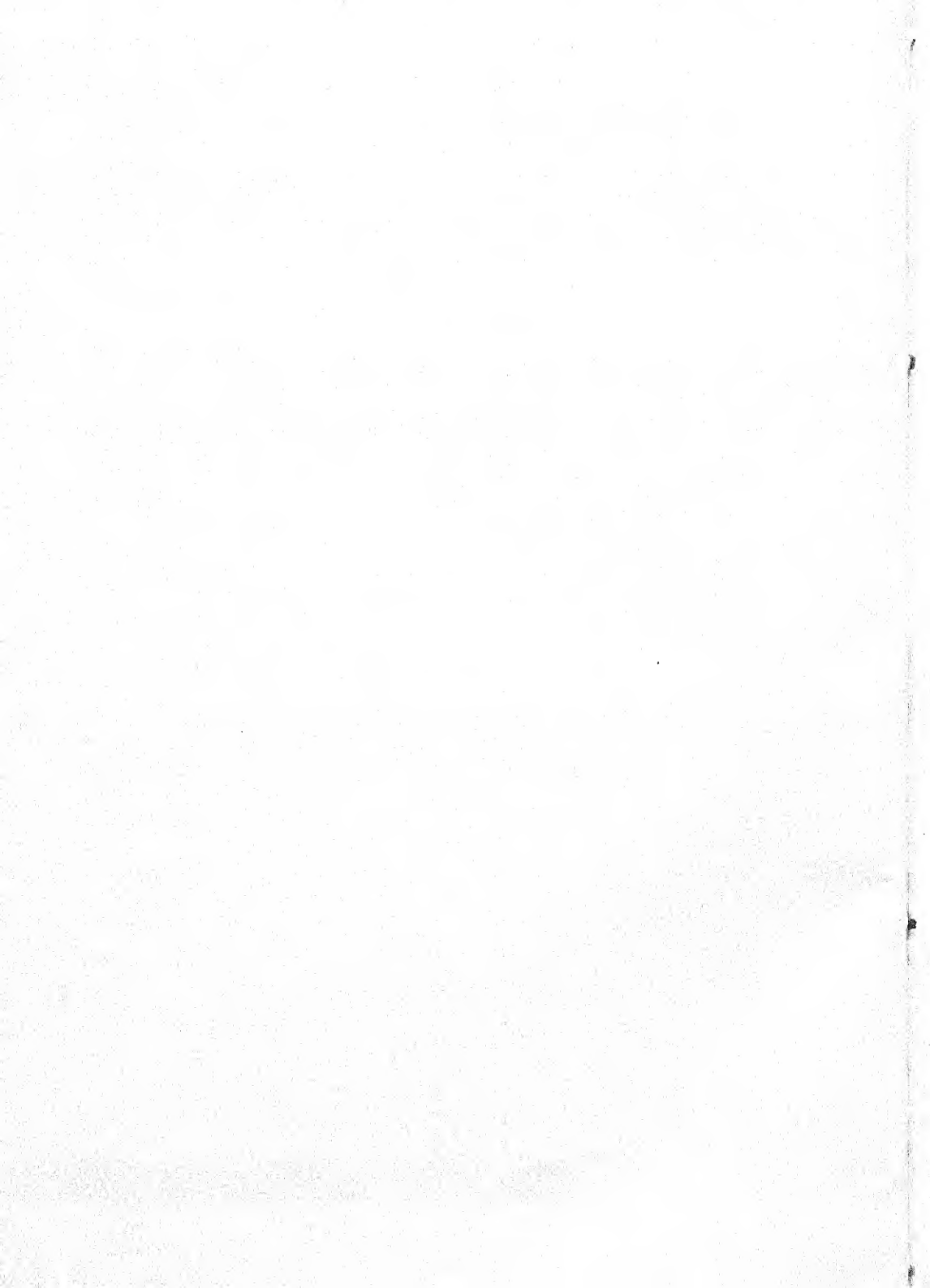
A 4-year crop rotation which included lespedeza gave better aggregation on plots 5 and 6 of the Conservation Station soil than did a continuous sod of shallow-rooted grasses.

Continuous cultivation of cotton on plot 10 of the Conservation Station resulted in a significant decrease in aggregation.

REFERENCES

- (1) BAVER, L. D. 1929 The effect of the amount and nature of exchangeable cations on the structure of a colloidal clay. *Missouri Agr. Exp. Sta. Res. Bul.* 129.
- (2) BAVER, L. D. 1935 Factors contributing to the genesis of soil micro-structure. *Amer. Soil Survey Assoc. Bul.* 16: 55-56.
- (3) BAVER, L. D. 1936 Aggregation of soils and calcium ion saturation. *Amer. Soil Survey Assoc. Bul.* 17: 28-30.
- (4) BAVER, L. D., AND HALL, N. S. 1937 Colloidal properties of soil organic matter. *Missouri Agr. Exp. Sta. Res. Bul.* 267.
- (5) BAVER, L. D., AND RHOADES, H. F. 1932 Aggregate analysis as an aid in the study of soil structure relationships. *Jour. Amer. Soc. Agron.* 24: 920-930.
- (6) BRADFELD, R. 1936 The value and limitation of calcium in soil structure. *Amer. Soil. Survey Assoc. Bul.* 17: 31-32.
- (7) BROWNING, G. M. 1937 Changes in the erodibility of soil brought about by the application of organic matter. *Proc. Soil Sci. Soc. Amer.* 2: 85-96.
- (8) DISEKER, E. G., AND YODER, R. E. 1936 Sheet erosion studies on Cecil clay. *Ala. Agr. Exp. Sta. Bul.* 245.
- (9) HARPER, H. J. 1937 Soil structure and moisture movement. *Proc. Soil Sci. Soc. Amer.* 2: 15-20.
- (10) JENNY, H. 1933 Soil fertility losses under Missouri conditions. *Missouri Agr. Exp. Sta. Bul.* 324.
- (11) LUTZ, J. F. 1934 The physico-chemical properties of soils affecting soil erosion. *Missouri Agr. Exp. Sta. Res. Bul.* 212.
- (12) LUTZ, J. F. 1936 The relation of the calcium and hydrogen ions to some physico-chemical properties of clays. *Amer. Soil Survey Assoc. Bul.* 17: 24-27.

- (13) LUTZ, J. F. 1937 The relation of free iron in the soil to aggregation. *Proc. Soil Sci. Soc. Amer.* 1: 43-45.
- (14) MERKLE, F. G. 1934 Base exchange studies on the Pennsylvania Jordan field plots. *Soil Sci.* 38: 37-64.
- (15) MYERS, H. E. 1937 Physicochemical reactions between organic and inorganic soil colloids as related to aggregate formation. *Soil Sci.* 44: 331-359.
- (16) PEELE, T. C. 1936 The effect of calcium on the erodibility of soils. *Proc. Soil Sci. Soc. Amer.* 1: 47-58.
- (17) PEELE, T. C. 1937 The effect of lime and organic matter on the erodibility of Cecil clay. *Proc. Soil Sci. Soc. Amer.* 2: 70-84.
- (18) PEELE, T. C. 1937 The relation of certain physical characteristics to the erodibility of soils. *Proc. Soil Sci. Soc. Amer.* 2: 97-100.
- (19) PRINCE, A. L., AND TOTH, S. J. 1936 The effect of phosphates on the cation exchange capacity of certain soils. *Soil Sci.* 42: 281-290.
- (20) TIURIN, I. V. 1931 New modification of the volumetric method of determining soil organic matter by means of chromic acid. *Pedology* 26: 36-47.
- (21) TOTH, S. J. 1937 Anion adsorption by soil colloids in relation to changes in free iron oxides. *Soil Sci.* 44: 299-314.
- (22) WAKSMAN, S. A. 1936 Humus. Williams & Wilkins, Baltimore, Md.
- (23) YODER, R. G. 1936 A direct method of aggregate analysis of soils and a study of the physical nature of erosion losses. *Jour. Amer. Soc. Agron.* 28: 337-351.



RATE OF PENETRATION OF LIME IN SOILS UNDER PERMANENT GRASS¹

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The production and maintenance of permanent grassland presents a somewhat different problem from ordinary field crop production, as regards liming and fertilization. For field crops, applications of fertilizers and lime can usually be thoroughly mixed with the soil by tillage operations, but this is not possible on permanently grassed areas. All materials which are applied to grassland must be spread on the surface. They penetrate the soil very slowly, unless artificial watering is practiced (13). Only a small percentage of turfed areas, however, receive artificial watering. It is with the areas which receive only natural rainfall that we are primarily concerned.

The results of pasture and turf studies in many parts of the world indicate that soil acidity and a low content of soil calcium are often important limiting factors in grass production (1, 5, 7, 9, 14). Since lime must be spread on the surface of the soil, penetration is slow, and the expected improvement does not always materialize.

Stewart and Wyatt (15) found that, after 14 years, only one-half of the acidity in the subsurface was neutralized where applications of lime had been heavy, and only one-fourth, where light. From the results of investigations in Kansas, on a series of alfalfa fertility plots, Metzger (10) concluded that surface-applied lime had penetrated to a depth of only about 6 inches during a 14-year period.

The results of Weidman (16) and Wilson (17) indicate that lateral and upward movement of all forms of lime is almost negligible, even with heavy applications. Frear (6), from studies made on bare fallow soil and on a closely trimmed sod in lysimeters, reports that, at the end of 1 year, half of the slaked lime and finely ground limestone remained where it had been placed, and only one-fourth of it had moved downward more than 1 inch.

In recent studies of the lime content and pH value of treated and untreated pasture soils, Nelson (12) found that of an application of 2400 pounds of limestone per acre none had penetrated beyond 6 inches and very little below 3 inches, during a 6-year period. Brown and Slate (5) reported that an application of 1 ton of limestone per acre to a permanent pasture had not affected the

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soil reaction below a 2-inch depth after 2 years. Brown and Munsell (2, 3, 4) studied the penetration of limestone in Connecticut pastures on Charlton fine sandy loam. With 2 tons per acre applied at 5-year intervals, maximum increase in pH was obtained 4 years after the second application, and penetration could still be observed 9 years after the second application. The pH remained constant in the surface 2 inches during this period. This indicates that more of the limestone was decomposing and moving downward, and not that the lower depths were becoming enriched with bases at the expense of the upper. They concluded that "10 years are necessary, following a two-ton application of limestone, to neutralize the acidity in the upper six inches."

EXPERIMENTAL PROCEDURE

This investigation was conducted to study the rate of penetration of surface applications to turf of finely ground limestone and hydrated lime. For this purpose, 12 soil types common to New Jersey were excavated to a depth of 4 inches, transported to the experimental area, and placed on plots that were 10 feet square, to a depth of 4 inches. (The entire plot area is underlain by Penn loam subsoil.) The plots were then sown to Kentucky bluegrass and red fescue. From 1929 forward, each plot received two applications yearly of an 8-6-4 fertilizer, each at the rate of 10 pounds per 1,000 square feet of area. In May, 1935, half of each plot received hydrated lime at the rate of 1 ton per acre. In November, 1938, an application of finely ground limestone, at the same rate, was made to a portion of each plot. One-fourth of each plot received no lime, one-fourth was limed in 1935, and one-fourth in both 1935 and 1938.

In November, 1935, and in March, June, August, and November, 1936, November, 1937, and November, 1939, samples were taken from the limed and unlimed portions of each plot. Twelve soil cores, each one-half inch in diameter, were taken from each plot to a depth of 4 inches on each date. These were cut into 1-inch sections, and composite samples were made from the corresponding sections. The surface quarter-inch of each soil core was discarded to eliminate all liming material that still remained on the surface. The samples were air-dried, crushed, passed through a 2-mm. screen, and tested for pH and available calcium. The data for the duplicate plots were averaged and are presented in the graphs and tables.

The pH values were determined electrometrically by the quinhydrone procedure. A modification of Morgan's (11) method was used to determine the content of available calcium. The buffered solution of sodium acetate employed in this method for extracting available plant nutrients has been found applicable to grassland soils by Grau (8). It was considered essential to have a rapid method of analysis for convenience in testing the large number of soil samples which were taken. Morgan's method was modified to the extent that instead of using 10 drops of extract for the available calcium determination, 0.5 cc. was measured out with 5-cc. graduated pipettes.

On December 11, 1939, and again on June 27, 1940, five soil cores, each 1 inch in diameter, were taken from each quarter of each plot. The surface mat of undecomposed clippings and roots was removed, and moisture determinations were made. Observations were made also on the vigor and sod density of each plot. The data from these determinations and observations are recorded in tables 2 and 3.

RESULTS

The 1-ton application of hydrated lime per acre made in May 1935 had a fairly constant effect upon the surface inch of all soil types, as shown in figures 1 and 2. The soil reaction was raised approximately one pH unit. In most cases, the maximum increase was obtained 6 months after application, and this reaction was maintained for the next 48 months. The additional ton of finely ground limestone, applied 42 months after the initial hydrated-lime treatment, effected a further increase, in all soil types, of approximately 0.5 pH.

Changes in pH below the first inch

The graphs obtained when the pH values of the second, third, and fourth inches were plotted are quite different from those of the first inch. One general type of curve was found in all soil types. This consisted of a slight rise in pH values at all three depths, over a period of 18 months. The magnitude of these increases was very low, usually from 0.1 to 0.3 pH unit, and in no case exceeded 0.6. The coarser-textured soils, such as Sassafras loamy sand and sandy loam, and Merrimac sandy loam, showed the greatest increase in pH below the first inch. Even in these soils, however, the pH change below the second inch was almost negligible, up to 18 months.

Between the 18- and 30-month periods there was a slight drop in pH at all depths below the first inch. This drop was characteristic of all soil types, but was much more noticeable on those types which had the greatest increase in pH up to 18 months. This decrease was probably caused by climatic factors which were operating to reduce the soil reaction during the fall of 1937. The rainfall records, shown in table 4, do not, however, adequately explain this phenomenon. There is a possibility that the large number of heavy showers during the 12-month interval, when this reduction in pH occurred, resulted in excessive leaching of bases from the horizons below the first inch. During this interval there was a total of 19 showers of 1 or more inch precipitation each, as compared with only 11, 10, and 12, during the other 12-month intervals. Some other climatic factor than rainfall, however, must also have been having an effect, but the weather records are not sufficient to offer an explanation.

During the 24-month interval between the 30- and 54-month periods there was an appreciable increase in pH in the second-, third-, and fourth-inch horizons. In some of the more porous soils, such as those of the Sassafras series and Merrimac sandy loam, the pH values of the second, third, and fourth

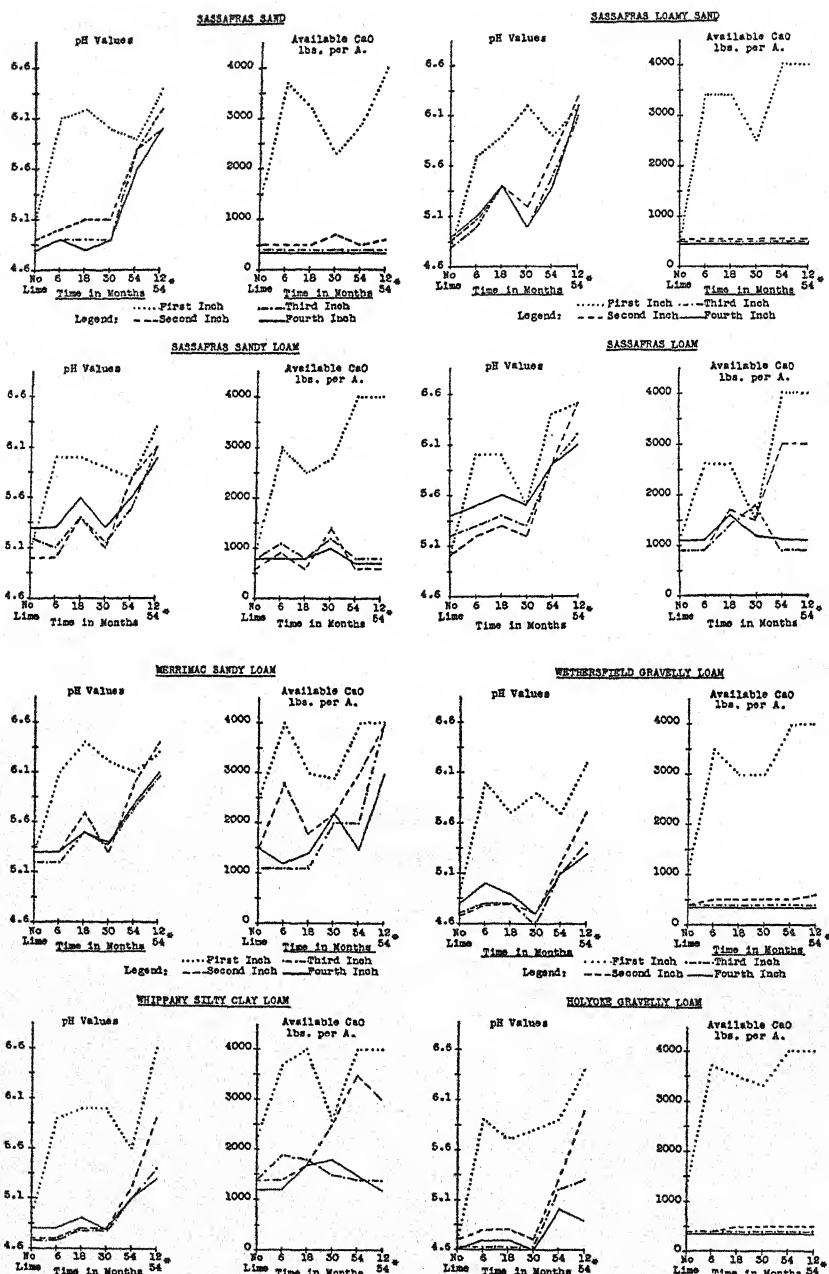


FIG. 1. CHANGES IN pH AND AVAILABLE CaO AT DIFFERENT DEPTHS IN VARIOUS SOIL TYPES AT 6-, 18-, 30-, AND 54-MONTH INTERVALS AFTER APPLICATION OF 1 TON PER ACRE OF HYDRATED LIME, AND 12 MONTHS AFTER AN ADDITIONAL 1-TON APPLICATION OF FINELY GROUND LIMESTONE SPREAD 42 MONTHS AFTER FIRST APPLICATION

* Twelve months after second and 54 months after first application of lime.

inches were almost identical with that of the first inch. The less fertile soil types, with less porous structures, such as Whippany silty clay loam, Wethers-

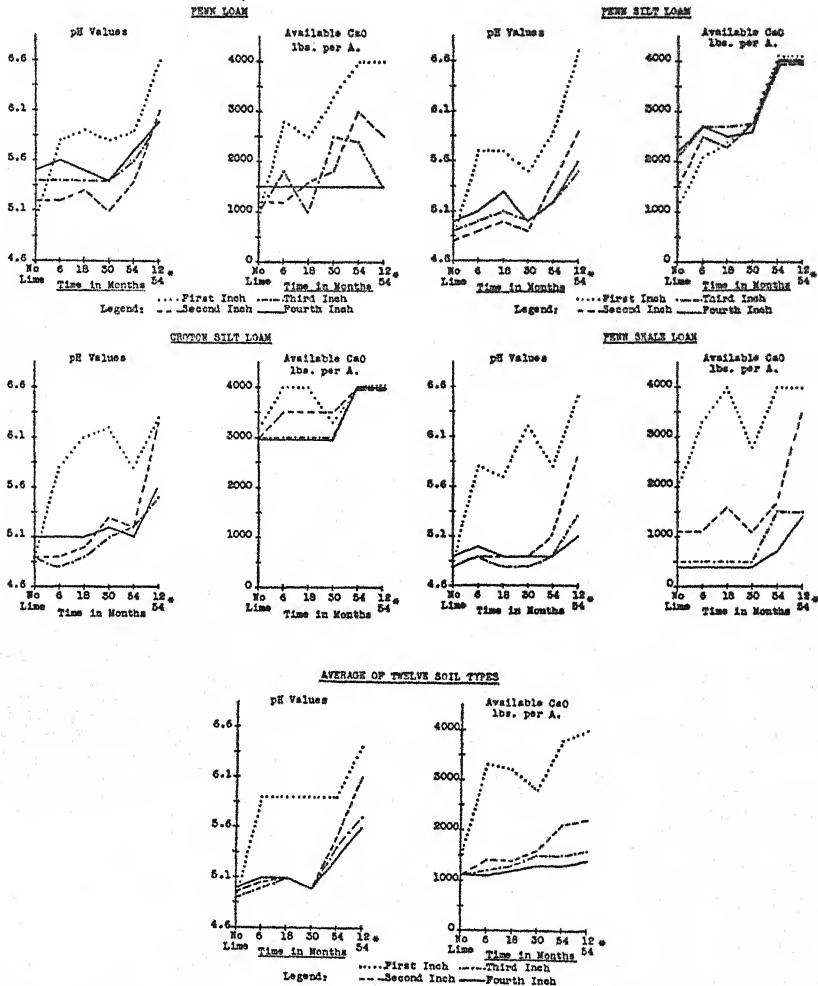


FIG. 2. CHANGES IN pH AND AVAILABLE CaO AT DIFFERENT DEPTHS IN VARIOUS SOIL TYPES AT 6-, 18-, 30-, AND 54-MONTH INTERVALS AFTER APPLICATION OF 1 TON PER ACRE OF HYDRATED LIME, AND 12 MONTHS AFTER AN ADDITIONAL 1-TON APPLICATION OF FINELY GROUND LIMESTONE SPREAD 42 MONTHS AFTER FIRST APPLICATION

* Twelve months after second and 54 months after first application of lime.

field and Holyoke gravelly loams, and Penn shale loam, had no significant increases in pH below the first inch.

The additional application of 1 ton per acre of finely ground limestone, made after a 42-month period, seemed to penetrate more rapidly than the first, but

the increase in pH in each soil type was closely correlated with increases obtained from the first application. That is, the soil types which had the greatest reduction in acidity from the first application of lime also had the greatest from the second. Only small increases in pH occurred below the second inch in the Whippany, Wethersfield, Holyoke, and Penn soils.

Changes in available calcium below the first inch

The changes in the available calcium content of several of the soil types could not be correlated with the pH changes, except in the first inch. The available calcium in the first inch increased in all soil types up to 6 months, and then decreased up to 30 months. From 30 to 54 months, the available calcium content increased again, and the second application of lime resulted in an excess of available calcium in the first inch.

Three groups of soils could be distinguished with regard to the rate of penetration below the first inch, as evidenced from the available calcium content. In group 1 there was no increase in available calcium below the first inch. In group 2, there was an increase in the second inch only. In group 3, there was an increase at all depths.

The soil types included in the first group were Sassafras sand, sandy loam, and loamy sand and Holyoke and Wethersfield gravelly loams. These soil types contained extremely low amounts of available calcium before lime applications were made, and no effects of either application could be noted below the first inch, with the analytical methods employed. This was true even though the Sassafras soils in this class had substantial increases in pH during the same period. This would seem to indicate that these soils had a low exchange capacity and were able to hold only small amounts of the soluble calcium. Since there was no increase in pH or in available calcium below the first inch in the Wethersfield and Holyoke gravelly loams, either these soils must have a high fixing power for calcium in the first inch, or the lime was held in the surface layer because of the low permeability of these soil types to water.

The soil types which had increases in available calcium only in the first and second inches included Whippany silty clay loam, Sassafras loam, and Penn loam. These soils were found to contain larger amounts of available calcium than those in group 1, even before lime was applied. The increases found in the second inch were too variable, however, to be correlated in any way with the increases in pH values.

The soil types which had increases in available calcium at all depths included Penn shale and silt loams, Merrimac sandy loam, and Croton silt loam. A much better correlation could be found between increases in pH and available calcium in these soils. Increases in pH values were almost always accompanied by increases in available calcium. The soils in this group were also found to be much higher in their content of available calcium than those in group 1, even before any lime applications were made. This would seem to indicate that the soils in this group had their exchange complexes fairly well

saturated with soluble calcium before any lime was applied. Apparently lime penetrated these soils readily and was held in the exchange complex.

Relative penetration of hydrated lime and ground limestone

Table 1 shows a comparison of hydrated lime, applied in the spring, with finely ground limestone, applied in the fall, in effect on pH. The pH values were obtained from samples taken from those quarter portions of each plot which had received only a single application of lime. The difference between these quarter plots was that one received an application of 1 ton per acre of

TABLE 1

Comparison of the pH values obtained in 18 months from 1 ton per acre of hydrated lime applied in May, 1935, with those obtained in 12 months from 1 ton of finely ground limestone applied in November, 1938

SOIL TYPE	pH AT DIFFERENT DEPTHS, 18 AND 12 MONTHS RESPECTIVELY, AFTER LIME APPLICATIONS								pH OF UNTREATED SOIL
	First inch		Second inch		Third inch		Fourth inch		
	Ca(OH) ₂ 18 Months	CaCO ₃ 12 Months	Ca(OH) ₂ 18 Months	CaCO ₃ 12 Months	Ca(OH) ₂ 18 Months	CaCO ₃ 12 Months	Ca(OH) ₂ 18 Months	CaCO ₃ 12 Months	
Sassafras sand.	6.2	6.0	5.1	5.2	4.9	5.0	4.8	4.9	4.9
Sassafras sandy loam. . .	6.0	5.8	5.4	5.3	5.4	5.4	5.4	5.4	5.2
Sassafras loamy sand. . .	5.9	6.1	5.4	5.5	5.4	5.6	5.4	5.5	4.9
Sassafras loam.	6.0	6.0	5.3	5.5	5.4	5.5	5.5	5.5	5.2
Merrimac sandy loam . . .	6.4	6.0	5.7	5.5	5.5	5.6	5.4	5.6	5.3
Whippany silty clay loam	6.0	5.9	4.8	4.8	4.9	4.9	4.8	4.8	4.8
Wethersfield gravelly loam.	5.7	5.9	4.8	5.0	4.8	4.9	4.8	4.8	4.8
Holyoke gravelly loam..	5.7	5.8	4.8	4.9	4.6	4.9	4.7	4.7	4.7
Penn loam.	5.9	6.0	5.3	5.3	5.4	5.4	5.5	5.4	5.3
Croton silt loam.	6.1	6.0	5.0	5.1	4.9	5.1	5.0	5.0	5.0
Penn silt loam.	5.7	5.7	5.0	5.1	5.1	5.1	5.1	5.0	4.9
Penn shale loam.	5.7	6.1	4.9	4.9	4.8	4.9	4.9	4.9	4.8
Average.	5.9	5.9	5.1	5.2	5.1	5.2	5.1	5.1	5.0

hydrated lime in May, 1935, and the pH values were determined 18 months later, whereas the other received an application of 1 ton per acre of finely ground limestone in November, 1938, and the pH values were determined 12 months later. The 18-month effect of hydrated lime was compared with the 12-month effect of finely ground limestone, because it seemed advisable to obtain all soil samples in the late fall under conditions which were as uniform as possible.

The values shown in table 1 indicate that hydrated lime, applied in spring, is less effective than finely ground limestone, applied in fall, even though the hydrated lime had been on 6 months longer. This was especially noticeable

in the second- and third-inch depths. At these depths the finely ground limestone produced a greater increase in the pH values than did hydrated lime, in all but four soil types. No difference could be noted between the effects of the two liming materials upon the pH values of the first and fourth inches.

Hydrated lime caked and hardened on the soil surface and remained resistant to the solvent effect of water for several months. Finely ground limestone (80 per cent passing a 100-mesh screen) did not cake and seemed to be more easily worked into the soil by natural agencies.

TABLE 2

Comparison of the effect of 1 ton of hydrated lime applied in May, 1935, with that of 1 ton of finely ground limestone applied in November, 1938, upon the moisture content, vigor, and density of red fescue and Kentucky bluegrass sod

SOIL TYPE	TYPE OF TURF*	MOISTURE CONTENT, 6/27/40		GRASS VIGOR INDEX,† 6/27/40		SOD DENSITY, 6/27/40	
		Limed 1935	Limed 1938	Limed 1935	Limed 1938	Limed 1935	Limed 1938
		per cent	per cent			per cent	per cent
Sassafras sand.	Fescue	5.4	4.5	2	3	95	85
Sassafras sandy loam.	Bluegrass	10.3	9.3	2.5	3	96	92
Sassafras loamy sand.	Fescue	11.6	6.5	2	3	98	94
Sassafras loam.	Bluegrass	14.8	16.5	2.5	2.5	96	96
Merrimac sandy loam.	Bluegrass	14.3	15.1	2	2.5	98	96
Whippany silty clay loam. .	Bluegrass and fescue	15.1	13.3	2	2	98	98
Wethersfield gravelly loam. .	Fescue and bluegrass	15.9	10.1	2.5	3	98	96
Holyoke gravelly loam.	Fescue and bluegrass	12.0	9.9	2.5	2	95	95
Penn loam.	Bluegrass and fescue	14.4	16.3	2.5	2	96	96
Croton silt loam.	Bluegrass and fescue	16.7	13.8	2	2.5	96	96
Penn silt loam.	Bluegrass	16.9	16.7	2	2	96	96
Penn shale loam.	Fescue and bluegrass	15.3	13.1	2.5	2.5	92	90
Average.		13.6	12.1	2.3	2.5	96	94

* The original seed mixture contained both fescue and Kentucky bluegrass.

† Vigor index: 1 = very good; 2 = good; 3 = medium; 4 = poor; 5 = very poor.

It was much easier to force the sampling tool into the limed portions than into the unlimed portion of each plot. For this reason, moisture determinations were made on all four quarters of each plot to a 4-inch depth. The results of these are shown in tables 2 and 3.

Definite differences could be noted in the vigor and density of the turf in the limed and unlimed portions of each plot during the dry season of 1939. Even with the above-normal rainfall during March, April, May, and June of 1940, these differences in soil moisture content could still be observed. Accordingly, observations were made on the vigor and density of each quarter plot. These observations are recorded in tables 2 and 3.

In table 2 are shown the determinations of soil moisture content and ob-

servations on the vigor and density of the sod on the quarter plots limed in May, 1935, and November, 1938. These observations were made on June 27, 1940, following an extremely wet spring. From March 1 to June 27, the precipitation was 7.2 inches more than the normal for that period (table 4). In the 3 days preceding the taking of the soil samples for moisture determinations, 2.04 inches of rain fell. Thus the differences in soil moisture content

TABLE 3

Effect of 1 ton of hydrated lime applied in May, 1935, and 1 ton of finely ground limestone applied in November, 1938, upon the moisture content, vigor, and density of red fescue and Kentucky bluegrass sod

SOIL TYPE	TYPE OF TURF*	MOISTURE CONTENT				GRASS VIGOR INDEX,†		SOD DENSITY, 6/27/40	
		12/11/39		6/27/40		6/27/40			
		No lime	Limed 1935, 1938	No lime	Limed 1935, 1938	No lime	Limed 1935, 1938	No lime	Limed 1935, 1938
		per cent	per cent	per cent	per cent			per cent	per cent
Sassafras sand.	Fescue	2.6	14.0	2.7	5.7	4.0	2.0	80	96
Sassafras sandy loam. ...	Bluegrass	3.7	8.3	5.8	11.9	3.5	2.5	90	95
Sassafras loamy sand.	Fescue	5.0	6.6	4.2	8.9	3	2	90	98
Sassafras loam.	Bluegrass	8.8	16.1	7.8	17.1	3	2	94	98
Merrimac sandy loam ...	Bluegrass	10.6	13.7	10.0	16.7	3	2	95	98
Whippany silty clay loam	Bluegrass and fescue	9.0	11.2	9.9	12.5	2	2	86	98
Wethersfield gravelly loam.	Fescue and bluegrass	7.4	13.0	9.9	15.2	4	2	90	98
Holyoke gravelly loam. ...	Fescue and bluegrass	7.4	11.5	8.4	15.5	4	2	85	96
Penn loam.	Bluegrass and fescue	10.9	13.7	11.2	15.7	2.5	2	96	98
Croton silt loam.	Bluegrass and fescue	13.6	18.7	11.0	10.9	3	2	92	96
Penn silt loam.	Bluegrass	16.7	18.9	12.7	17.0	2.5	2	94	96
Penn shale loam.	Fescue and bluegrass	13.8	14.6	10.9	15.6	4.0	2.5	87	95
Average.		9.1	13.4	8.7	13.6	3.3	2.1	90	97

* The original seed mixture contained both fescue and Kentucky bluegrass.

† Vigor index: 1 = very good; 2 = good; 3 = medium; 4 = poor; 5 = very poor.

were primarily due to the ability of each differently treated soil to absorb water.

The figures for moisture content, shown in table 2, indicate that the 1935 lime application made the soil slightly more permeable to water than the 1938 application. In all but three soil types, the moisture content was higher where lime had been applied in 1935. The average moisture content for all soil types was 13.6 per cent, for those limed in 1935, as compared with 12.1 per cent, for those limed in 1938. When the latter value, however, was compared with 8.7 per cent (the average moisture content for the unlimed soils as shown in table 3), it was readily seen that the lime applied in November,

1938, had effected a substantial improvement in the permeability of the soil to water, even though the lime had been on the soil only 18 months. It was also found that the single application of lime, in 1935, had been as effective as the two applications of 1935 and 1938, in increasing the porosity of the soil, judging by the identical average moisture contents of 13.6 per cent (tables 2 and 3).

Moisture determinations were also made on samples from the unlimed and double-limed portions of each plot on December 11, 1938. These results are shown in table 3. Even though these determinations were made following a 2-month period in which the rainfall was 1.4 inches less than normal (table 4), the moisture contents in both cases were almost identical with those of samples collected in the spring, following a period of excessive rainfall.

The effects of the lime could also be observed in the vigor and density of the sod, as shown in tables 2 and 3. These observations show that the portion

TABLE 4

Precipitation by intervals covering the period of the lime penetration studies, June 1, 1935 to June 27, 1940

PERIOD	INTERVAL	PRECIPITATION	DEVIATION FROM NORMAL	NUMBER OF SHOWERS OVER 1 INCH
	<i>months</i>	<i>inches</i>	<i>inches</i>	
June 1, 1935, to December 1, 1935.....	6	27.2	+5.1	9
December 1, 1935, to December 1, 1936.....	12	40.6	-4.9	11
June 1, 1935, to December 1, 1936.....	18	67.8	+ .3	20
December 1, 1936, to December 1, 1937.....	12	48.5	+3.0	19
December 1, 1937, to December 1, 1939.....	24	95.2	+4.2	22
December 1, 1938, to December 1, 1939.....	12	38.5	-7.0	12
October 11, 1939, to December 11, 1939.....	2	5.0	-1.4	
March 1, 1940, to June 27, 1940.....	4	21.8	+7.2	

of the plot which received lime in both 1935 and 1938 had a more vigorous, dense sod than either of the portions receiving single applications of lime in 1935 and 1938. The 1938 application, however, materially improved the vigor and density of the turf above that where no lime was applied. The average vigor indexes for all soil types were 3.3 for unlimed; 2.5 and 2.3, respectively, for the 1938 and 1935 lime applications; and 2.1 where lime was applied in both 1935 and 1938 (1.0 indicates great vigor, 3.0 medium vigor). This same order of improvement was also found in the density of the sod. The average sod density for all soil types was: unlimed, 90 per cent; limed 1938, 94 per cent; limed 1935, 96 per cent; and limed in both 1935 and 1938, 97 per cent, the percentage values indicating the degree of occupation of the soil surface by the grass sod.

SUMMARY AND CONCLUSION

The effect of surface applications of hydrated lime and ground limestone on established sods on 12 soil types which had all been moved to one location,

was studied with respect to rate of changes in soil pH values, associated changes in available calcium, permeability as measured by soil moisture content, and response of the grass.

There was a sharp rise in pH of soil in the upper inch on all soil types by the end of 6 months, but substantial changes at 2-, 3-, and 4-inch depths did not occur until after 30 months had elapsed. A second lime application, made 42 months after the first, produced a sharp increase in pH of the 2-, 3- and 4-inch layers, within a 12-month period.

One ton of finely ground limestone, applied in November, 1938, produced as great an effect on the pH below the upper inch, by the end of 12 months, as 1 ton of hydrated lime applied in May, 1935, after an 18-month period. The hydrated lime became caked and dissolved more slowly than the limestone, which remained in a finely divided condition irrespective of moisture conditions.

Changes in available calcium, after liming, varied with the soil type. Soils of group 1, represented by Sassafras sand, sandy loam, and loamy sand and Wethersfield and Holyoke gravelly loams, showed no increase in calcium below the upper inch. The first two soils apparently had low exchange capacity and retained but little of the calcium which had passed through them. The Holyoke and Wethersfield soils had low calcium at the beginning, as well as low permeability to water, and this reduced the penetration of lime. Group 2, consisting of Whippany silty clay loam, Sassafras loam, and Penn loam, having a higher initial lime content than the soils of group 1, showed an increase in available calcium in the upper 2 inches, following liming. Soils of group 3, comprising Penn shale and silt loams, Merrimac sandy loam, and Croton silt loam, showed increases in calcium at all depths, and these were correlated with pH changes. The initial content of calcium was comparatively high in these soils, and permeability to water was reasonably good under the conditions of this experiment.

The moisture content in the upper 4 inches of soil was greatly increased as a result of liming. The vigor and density of sod was also improved, but whether this was due to more abundant soil moisture or to changes in pH and available calcium, was not determined.

REFERENCES

- (1) BENDER, C. B. 1934 Intensive pasture management. N. J. Agr. Exp. Sta. Bul. 623.
- (2) BROWN, B. A., AND MUNSELL, R. I. 1933 Penetration of surface applied lime and phosphates in the soil of permanent pastures. Conn. (Storrs) Agr. Exp. Sta. Bul. 186.
- (3) BROWN, B. A., AND MUNSELL, R. I. 1936 Soil reaction at various depths as influenced by the time since application, placement and amount of limestone. *Proc. Soil Sci. Soc. Amer.* 1: 271-275.
- (4) BROWN, B. A., AND MUNSELL, R. I. 1938 Soil acidity at various depths as influenced by time since application, placement, and amount of limestone. *Proc. Soil Sci. Soc. Amer.* 3: 217-222.
- (5) BROWN, B. A., AND SLATE, W. L. 1929 The maintenance and improvement of permanent pastures. Conn. (Storrs) Agr. Exp. Sta. Bul. 155.

- (6) FREAR, W. 1921 Lime topdressing experiment. *Pa. Agr. Exp. Sta. Bul.* 170: 24.
- (7) GARDNER, F. D., ET AL. 1935 Pasture fertilization. *Pa. Agr. Exp. Sta. Bul.* 323.
- (8) GRAU, F. V. 1935 Permanent pastures in Maryland. *Md. Agr. Exp. Sta. Bul.* 373.
- (9) LEA, C. L., AND MIDGLEY, A. R. 1934 Available potash and phosphorus contents of Vermont pasture soils. *Vt. Agr. Exp. Sta. Bul.* 373.
- (10) METZGER, W. H. 1934 Distribution of fertilizer residues in the soil after fourteen years of fertilizer experiment with alfalfa. *Jour. Amer. Soc. Agron.* 26: 620-625.
- (11) MORGAN, M. F. 1937 The universal soil testing system. *Conn. Agr. Exp. Sta. Bul.* 392.
- (12) NELSON, P. R. 1929 Lime penetration resulting from surface application to pasture land. *Soil Sci.* 27: 143-146.
- (13) SPRAGUE, H. B. 1932 Root development of perennial grasses and its relation to soil conditions. *Soil Sci.* 36: 189-209.
- (14) SPRAGUE, H. B., BLASER, R. E., AND BOYCE, J. H. 1940 A comparison of treatments on permanent pastures. *N. J. Agr. Exp. Sta. Bul.* 673.
- (15) STEWART, R., AND WYATT, F. A. 1919 Limestone action on acid soils. *Ill. Agr. Exp. Sta. Bul.* 212: 265-295.
- (16) WEIDEMAN, A. O. 1936 Movement of lime in soils as determined by soil reaction. *Mich. State Quart. Bul.* 4: 254-259.
- (17) WILSON, B. D. 1918 The translation of calcium in soil. *N. Y. (Cornell) Agr. Exp. Sta. Mem.* 17: 289-300.

THE INFLUENCE OF TWO JUNIPERUS SPECIES ON SOIL REACTION¹

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That the soil reaction is influenced by vegetation is well recognized. Various European investigators (3, 4, 5, 8, 9, 12, 14, 15) have shown that both the composition of the forest stand and the cultural operations in the stand affect the pH of the soil.

It is reasonable to assume that a plant may influence the reaction of the soil beneath it by any or all of three means: first, by changing the chemical composition of the soil through withdrawals of substances by the roots; second, by changing the chemical composition of the soil through additions of substances from the decomposition of litter; and third, indirectly through modifying soil structure, shading the surface, intercepting rainfall, etc.

Kosłowska (6), working with 39 plant species, showed that "plants possess in their underground organs the property of changing the reaction of the medium." That the decomposition of litter affects soil reaction has been demonstrated and at least partly explained by Nemec and Kvapil (8, 9), Stepanof (12), Waksman (13), and others.

EXPERIMENTAL

In order to measure the influence of vegetation on soil reaction, experiments were carried on in the vicinity of New Haven in the fall and winter of 1939-40, Red cedar, *Juniperus virginiana*, and ground juniper, *Juniperus communis*, growing on old field soils were chosen for this purpose; and on three divisions of the Eli Whitney Forest of the New Haven Water Company, blocks were laid out, in each of which a series of random samples was taken. Four paired samples were taken at each location, two under the plant at depths of 1 inch and 6 inches in the mineral soil and two at a distance of 5 feet from the plant at the same depths. At the latter position, that is, in the open, the vegetation throughout consisted of pasture grasses.

The red cedars and one block of the ground junipers studied were growing on Gloucester sandy loam. The blocks on this brown glacial till soil, derived

¹ From a thesis submitted to the Yale School of Forestry in partial fulfillment of the requirements for the degree master of forestry. Acknowledgment is due for the assistance and advice given by H. J. Lutz throughout the work and by W. H. Meyer in the statistical analysis.

from granite and gneiss material, were on high, rocky, and well-drained land. The ground juniper was also studied on Cheshire sandy loam and on Holyoke loam (7). The block on Cheshire sandy loam, a soil derived from red Triassic sandstone, was situated on a moderate slope, low in a small stream valley but well drained and almost free from rocks, whereas that on Holyoke loam, derived from traprock, was near the top of a moderately high traprock ridge.

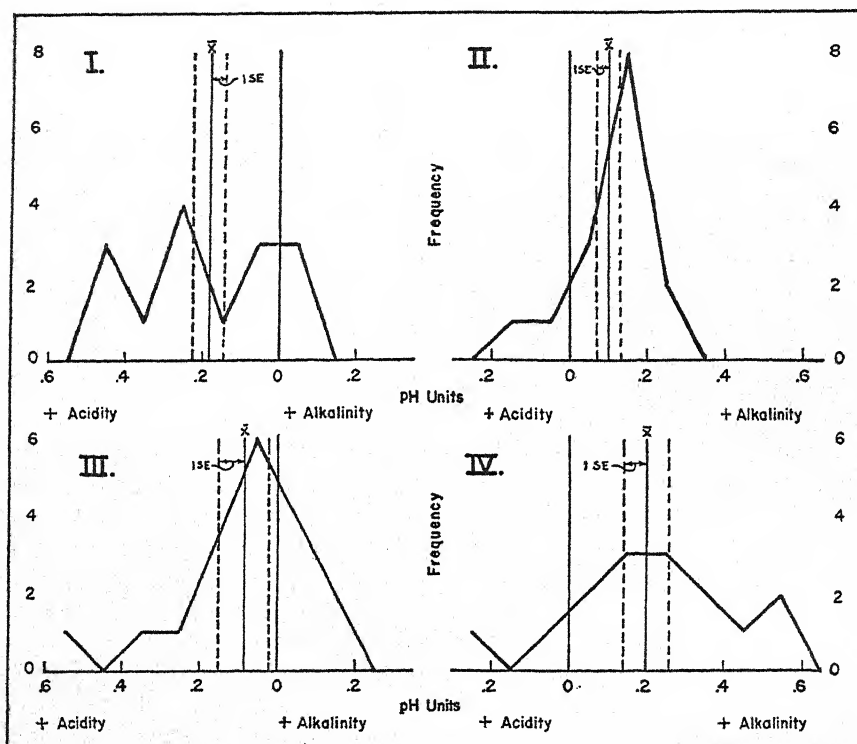


FIG. 1. COMPARISON OF pH VALUES FOR *JUNIPERUS VIRGINIANA* SAMPLES SHOWING FREQUENCY CURVES FOR EACH PAIR COMPARED, MEAN DIFFERENCES (\bar{X}), AND RANGE OF INDIVIDUALLY COMPUTED STANDARD ERRORS (SE)

- I. 6-inch depth under plant cf. with 1-inch depth under plant.
- II. 6-inch depth in open cf. with 1-inch depth in open.
- III. 1-inch depth in open cf. with 1-inch depth under plant.
- IV. 6-inch depth in open cf. with 6-inch depth under plant.

The effect of the two species on the soil profile was similar on all soil types. In contrast to the A₁ profile of the soil in the open, which tended to have little structure and to be compact and poorly aerated, the A₁ profile under the plants was characterized by a highly developed crumb structure coupled with considerable earthworm activity. Although a layer of raw humus was commonly found under other woody plants on the same soils, this layer was completely absent under the two *Juniperus* species. Below the A₁ horizon, there was no

observed difference between the soil profiles in the open and those under the canopy.

The soil samples taken were stored in paper bags until dry; pH determinations were then made by the quinhydrone method (16). In this determination, the apparatus was checked every half day with a standard buffered solution. The soil samples were found to be well suited for determination by this method, having small drifts and pH values between 4.4 and 5.6. The soil-liquid ratio

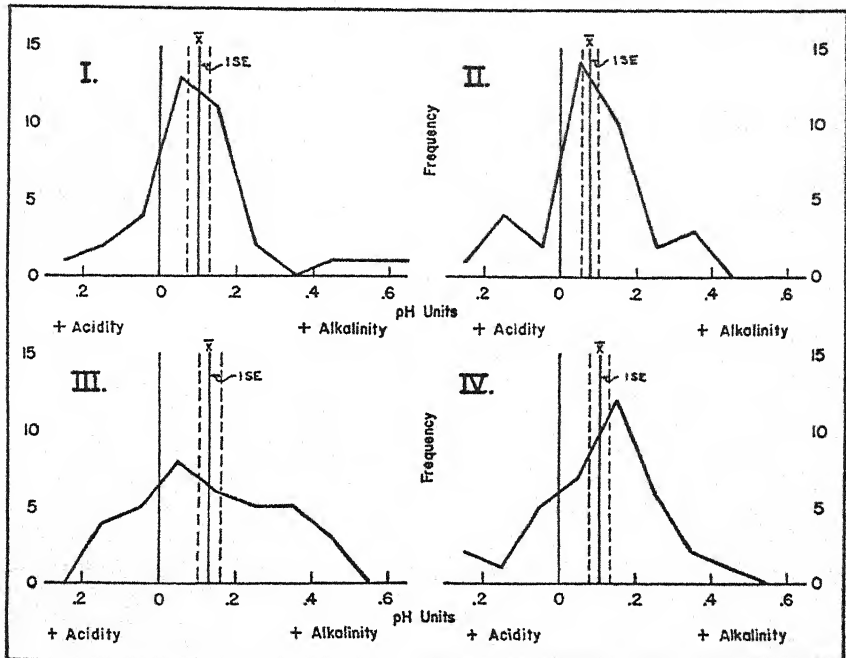


FIG. 2. COMPARISON OF pH VALUES FOR *JUNIPERUS COMMUNIS* SAMPLES SHOWING FREQUENCY CURVES FOR EACH PAIR COMPARED, MEAN DIFFERENCES (\bar{X}), AND RANGE OF INDIVIDUALLY COMPUTED STANDARD ERRORS (SE)

- I. 6-inch depth under plant cf. with 1-inch depth under plant.
- II. 6-inch depth in open cf. with 1-inch depth in open.
- III. 1-inch depth in open cf. with 1-inch depth under plant.
- IV. 6-inch depth in open cf. with 6-inch depth under plant.

used was that of 1:2.5. The soil and water were shaken together for approximately 30 seconds and the quinhydrone stirred in for 10 seconds. The potential was measured at 60 seconds.

A preliminary analysis of variance was made to determine whether the effect of ground juniper on the soil reaction varied with the soil type. A significant difference was found to exist between the blocks, but inasmuch as this significance was of a low order and the same general relationships were found to occur on each soil type, soil types were disregarded in subsequent analyses. Figures 1 and 2 show the distribution of differences of paired samples for each compar-

ison made. The standard errors of the difference indicated in these figures were derived separately for each comparison and not by analysis of variance as is done below.

In analyzing the data, analysis of variance was adopted, following the standard methods as laid down by Snedecor (11) and Paterson (10). The results are shown in tables 1 and 2. In the analysis of variance in table 2, the individual interactions were found to be of no significance and were grouped together to serve as the error.

TABLE 1
Juniperus virginiana: Analysis of variance

	SUM SQUARES	DF	VARIANCE	F	SIGNIFICANCE
Total.....	3.9988	59			
Location.....	2.6364	14	.1883	60.8	High
Position.....	.0510	1	.0510	16.5	High
Depth.....	.0244	1	.0244	8.1	Significant
Interactions					
Location and position.....	.7286	14	.0520	16.8	High
Location and depth.....	.2105	14	.0150	4.84	High
Position and depth.....	.3039	1	.3039	98.0	High
Location, position, and depth (error).....	.0440	14	.0031		

TABLE 2
Juniperus communis: Analysis of variance

	SUM SQUARES	DF	VARIANCE	F	SIGNIFICANCE
Total.....	9.480	143			
Location.....	7.190	35	.205	14.14	High
Position.....	.476	1	.476	32.83	High
Depth.....	.277	1	.277	19.10	High
Interactions (error).....	1.537	106	.0145		

The analysis of variance was followed by *t* tests comparing the average values for each position and depth (2). The results are presented in tables 3 and 4. The analysis showed a highly significant difference (*P* less than 0.01) in each case between the two depths at each position and between the two positions at each depth.

In the case of red cedar, measurements taken in the open showed the surface soil to be the more acid, the average pH at the 1-inch depth being 4.88 and that at the 6-inch depth, 4.98. Under the cedar, the pH gradient is reversed, the pH at the 1-inch depth being 4.96 and that at the 6-inch depth, 4.78. This is in agreement with the observation of Coile (1). The increase in the pH value of the surface soil under the cedar is clearly attributable to the incorporation of the decomposition products of litter. The pH of red cedar litter, as

determined from the plants studied, averaged 5.57. At the 6-inch depth under the cedar, a decrease in the soil pH as compared to that in the open occurred, a decrease very likely attributable to the withdrawal of calcium and other basic salts by the roots in that region. This decrease at the 6-inch depth was definitely greater than the increase at the 1-inch depth.

In the case of the juniper, the typical pH profile is again found in the open, the surface soil being the more acid. At the 1-inch depth, the pH was 5.05, and that at the 6-inch depth, 5.13. Under the juniper, a highly significant

TABLE 3

Juniperus virginiana: t-tests

AVERAGE VALUES COMPARED	DIFFERENCE (pH UNITS)	SE _{Diff.}	t	SIGNIFICANCE
1-inch depth under cedar and 6-inch depth under cedar1827	.00203	9.00	High
1-inch depth under cedar and 1-inch depth in open0840	.00203	4.13	High
6-inch depth under cedar and 6-inch depth in open2007	.00203	9.86	High
1-inch depth in open and 6-inch depth in open1020	.00203	5.02	High

TABLE 4

Juniperus communis: t-tests

AVERAGE VALUES COMPARED	DIFFERENCE (pH UNITS)	SE _{Diff.}	t	SIGNIFICANCE
1-inch depth under plant and 6-inch depth under plant099	.0284	3.48	High
1-inch depth under plant and 1-inch depth in open127	.0284	4.47	High
6-inch depth under plant and 6-inch depth in open103	.0284	3.63	High
1-inch depth in open and 6-inch depth in open076	.0284	2.68	High

drop in pH was observed at both depths, the pH at the 1-inch depth being 4.92 and that at the 6-inch depth being 5.02. At the surface, a striking similarity was noted between the pH of the litter, which was determined as 4.91, and that of the mineral soil, which was 4.92. Again, the changes that were found to have occurred under the plant can probably be attributed largely to the addition of litter at the 1-inch depth and to root action at the 6-inch depth.

SUMMARY

Both *Juniperus virginiana* and *Juniperus communis* alter the pH of old field soils in the vicinity of New Haven. The first species raises the pH of the upper part of the mineral soil and lowers it at a depth of 6 inches. *Juniperus*

communis, on the other hand, lowers the pH at both depths. Tentatively, it may be concluded that the addition of litter is a highly important factor influencing the pH of the upper part of mineral soil and withdrawal of soluble substances by the roots appears to be of similar importance at a 6-inch depth.

REFERENCES

- (1) COILE, T. S. 1933 Soil reaction and forest types in the Duke Forest. *Ecology* 14: 323-333.
- (2) FISHER, R. A. 1934 Statistical Methods for Research Workers, ed. 5. Oliver and Boyd, Edinburgh.
- (3) FRANK, E. 1927 Über Bodenazidität im Walde. Speyer und Kaerner, Freiburg im Breisgau.
- (4) HESS, E. 1929 Le sol et la forêt. *Mitt. Schweiz. Centralanst. Forstl. Versuchsw.* 15: 5-50.
- (5) HESSELMAN, H. 1937 Om humustäckets beroende av benståndets ålder och sammansättning i den nordiska granskogen av blåbärsrik *Vaccinium*-typ och dess inverkan på skogens föryngring och tillväxt. (Über die Abhängigkeit der Humusdecke von Alter und Zusammensetzung der Bestände im nordischen Fichtenwald von blaubeerreichem *Vaccinium*-Typ und über die Einwirkung der Humusdecke auf die Verjüngung und Wachstum des Waldes.) *Meddel. Statens Skogsför-söksanst.* 30: 529-716.
- (6) KOSLOWSKA, A. 1934 The influence of plants on the concentration of hydrogen ions in the medium. *Jour. Ecology* 22: 369-419.
- (7) MORGAN, M. F. 1939 The soil characteristics of Connecticut land types. Conn. Agr. Exp. Sta. Bul. 423.
- (8) NĚMEC, A., AND KVAPIL, K. 1925 Études sur les relations entre certaines propriétés physiques des sols forestiers et leur acidité. *Rev. Eaux et Forêts* 63: 393-406.
- (9) NĚMEC, A., AND KVAPIL, K. 1926 Studien über einige chemische Eigenschaften der Profile von Waldböden. *Ztschr. Forst u. Jagdw.* 58: 461-489, 525-554.
- (10) PATERSON, D. D. 1939 Statistical Technique in Agricultural Research. McGraw-Hill, New York.
- (11) SNEDECOR, G. W. 1937 Statistical Methods. Collegiate Press, Iowa.
- (12) STEPANOF, N. N. 1929 Chemical properties of the forest litter as fundamental factors in forest reproduction. Trudy Lesnomu Opytn. Dyelu, Moscow. (English translation furnished by H. J. Lutz.)
- (13) WAKSMAN, S. A. 1938 Humus, ed. 2. Williams & Wilkins, Baltimore.
- (14) WIEDEMANN, E. 1928 Untersuchungen über den Säuregrad des Waldbodens im sächsischen oberen Erzgebirge. *Ztschr. Forst u. Jagdw.* 60: 659-683.
- (15) WIEDEMANN, E. 1934 Über die Beziehungen des forstlichen Standortes zu dem Wachstum und dem Wirtschaftserfolg im Walde. *Deut. Forsch.* 24: 5-106.
- (16) WRIGHT, C. H. 1939 Soil Analysis: a Handbook of Physical and Chemical Methods, ed. 2. Thomas Murby and Co., London.

MICROBIAL ACTIVITIES IN SOIL: VI. MICROBIAL NUMBERS AND NATURE OF ORGANIC MATTER IN VARIOUS GENETIC SOIL TYPES¹

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Humus and its decomposition products are thought to be vital factors in the development of specific soil properties which differentiate soil types and the genetic horizons in soil profiles. Since humus is a product of organic matter transformation initiated and promoted by microbial action, the kind and activity of the microflora participating in this transformation should have a definite effect upon the nature of the humus and consequently the inherent soil properties.

Marked differences in both kinds and numbers of microbes in different genetic soil types and in different horizons of the same profile have been noted by various investigators. In a study of a number of genetic soil types in Iowa, Brown and Benton (1) found that, as a whole, microbial numbers decreased from the eluvial to the illuvial horizons. Soil type differences were reflected by differences in microbial numbers, which apparently were influenced to the greatest extent by soil organic matter content. The relationships between the numbers of bacteria, actinomycetes, and fungi were similar in different soil types in most cases. Razumov and Remezov (6) observed decreasing numbers of microbes with depth in the same horizon of podzol soils, and significant differences in microbial populations in the same genetic horizons of soils differing morphologically. Mal'chevkaia (3) who studied various podzol soils, found it difficult to ascribe to any soil type or to its genetic horizons any specific microbial character with respect to kind and numbers of microbes.

In several forest and grassland soils in Canada, Timonin (9) observed that in certain cases the B horizon supported a larger population of bacteria and actinomycetes than did the A horizon. The numbers of these organisms were from two to twenty times greater in the grassland than in the forest soils. Remezov (7) in a comprehensive review of the work on the carbon-nitrogen relationship of the organic matter in Russian soils contends that this relationship has a direct effect upon the intensity and kind of microbial activity and that the vegetation and kinds of soil microbes also influence the carbon-nitrogen relationship.

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² Professor of soils and formerly research assistant respectively. The authors gratefully acknowledge the assistance of L. E. Dunn, research assistant, who made the analysis of variance for the microbial numbers.

Although marked differences in numbers and kinds of soil microbes in different soil types have been noted, the cause of these variations has not been investigated thoroughly. Previous work in this laboratory (10, 11) indicates that the nature of the humus, as well as the nature of the organic residues available for humification, influences both the kind and activity of the soil microflora. The main object of the work presented in this paper was to study the numbers of organisms belonging to the principal groups of microbes participating in organic matter transformations in different genetic soil types and horizons, in an attempt to correlate microbial activity with variations in the amount and chemical composition of the soil humus.

EXPERIMENTAL PROCEDURE

Soil samples were obtained from the different horizons of 15 profiles representing a number of different genetic soil types. The samples were air dried and then passed through a 16-mesh sieve to effect thorough mixing and the removal of coarse organic debris. Portions of 2 kgm. of the air-dried, sifted soils were placed in 1-gallon glazed pots, and sufficient water was added to adjust the soil moisture content to the normal field moisture percentage, which was determined in advance on separate samples. The pots, covered loosely, were incubated at 28°C., and the soil moisture content was maintained within narrow limits throughout the experiment by additions of water when necessary.

The numbers of bacteria, actinomyces, fungi, aerobic cellulose-decomposing bacteria, and *Azotobacter* were determined at four 4-week intervals in 1937, after the soil in each pot was sifted twice through a 6-mesh sieve on clean paper in order to insure thorough mixing. Small samples of the sifted soil were taken for microbial counts, the procedures described in a previous paper (10) being employed.

Analyses for ammonia and nitrate-nitrogen were made at the beginning and at the end of the experiment. Further chemical determinations on the original soil samples consisted of pH values, oxidation-reduction potentials, total organic carbon, total nitrogen, and organic matter fractionations. Standard methods of analysis described previously (11) were used for all except the organic matter fractionations. The latter were made by the method of Waksman and Stevens (13) modified to the extent that the carbon in the extracts and residues was determined directly according to the procedure employed in previous work (11).

A general description of the origin and nature of the soils³ used in this experiment follows:

1. Ephrata sandy loam. This soil is a representative of the northern gray desert soils and covers extensive areas in central Washington. Its profile characteristics are described by Marbut (4).

³ Several of the genetic soil types represented in this study were supplied by colleagues from other states as follows: soil 2, W. T. McGeorge, University of Arizona; 4, E. H. Tyner,

2. Mohave clay loam. The Mohave series is representative of the southern gray desert soils described by Marbut (4).
3. Ritzville fine sandy loam. The Ritzville series covers large areas in central eastern Washington and closely resembles the brown soils. Its profile characteristics are described by Marbut (4).
4. Barnes clay loam. The Barnes series is representative of the northern chernozem soils described by Marbut (4).
5. Garrison gravelly loam. The Garrison soil, which resembles the northern chernozem type, is found in Spokane Valley, Washington. It has developed from glacial outwash and contains much gravel in the upper horizons. The lower horizons consist of a mixture of coarse gravel and cobbles.
6. Palouse silt loam. The Palouse series covers extensive areas in eastern Washington. The profile used in this study resembles that of the northern prairie soils; its characteristics are described in a previous publication (11).
7. Houston clay. The Houston series is representative of the rendzina soils as described by Marbut (4). The profile samples were obtained from Texas.
8. Spanaway gravelly sandy loam. This is a grassland soil found in limited areas in a forested region and developed from glacial outwash in the Tacoma area in Washington. Its profile characteristics, which are somewhat atypical of northern prairie soils, are described by Nikiforoff (5).
9. Spanaway gravelly sandy loam. This is a forest soil found adjacent to soil 8 and also developed from glacial outwash. It contains an unusually large amount of organic matter for a forest soil and occurs in the area of yellow-brown forest soils described by Nikiforoff (5).
10. Lynden sandy loam. The Lynden series is a typical representative of the yellow-brown forest soils in western Washington. Its profile characteristics are similar to those described by Wildermuth et al. (14).
11. Alderwood loam. The Alderwood series represents the same genetic type as the Lynden soil. Its profile characteristics are described in detail by Wildermuth et al. (14).
12. Helmer silt loam. This is a brown-colored forest soil developed on loessial material. Its profile description is given in a previous paper (11).
13. Rainier sandy loam. This soil, which has not been classified officially, is an immature podzol developed under the influence of vertical zonality on the northeast slopes of Mount Rainier 3 miles north of the Yakima Park entrance. The parent material is glacial outwash. The A₁ horizon, which overlies coarse gravel, is well developed, but there is no distinct A₂ horizon.
14. Lakewood sandy loam. The Lakewood soil is a representative of the podzol soils in New Jersey. The profile characteristics are described by Joffe (2).
15. Greenville fine sandy loam. The Greenville soil represents the red and yellow soils as described by Marbut (4).

The A₀ horizons or organic accumulative layers of the foregoing soils were not included in this study but were reserved for a separate investigation which will be reported in a subsequent paper.

EXPERIMENTAL RESULTS

It is generally recognized that the kind and activity of the microflora of soils exposed to identical temperature and moisture conditions are affected

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principally by various inherent soil properties such as texture, structure, and reaction, as well as by the nature and quantity of the available organic and inorganic food supply. The general and specific soil properties are broadly expressed by the character of the genetic soil type, and the food supply in the various soil profile horizons is influenced in large measure by the amount and chemical composition of the humus. The effect of genetic soil types and of the nature and quantity of soil humus on the kind and activity of the microflora in the 15 soil profiles included in this study will be considered separately.

Microbial numbers in various genetic soil types

The results of the four periodical microbial determinations are recorded in table 1, and the average counts are graphically illustrated in figure 1. Table 1 contains no data on *Azotobacter*, as these organisms were found only in small numbers in the A horizons of 4 of the 15 soil types. These 4 soils are the Helmer, Lynden, Barnes, and Houston series, which produced 5, 5, 10, and 280 colonies per gram of dry soil respectively.

A notable feature revealed by the data on microbial counts, and one that is not unusual in microbial determinations in soils, is the marked fluctuation in numbers of bacteria, fungi, actinomyces, and cellulose-decomposing bacteria at different periods of sampling. This occurred in spite of the fact that uniform temperatures and soil moisture contents were maintained during the incubation period of 108 days and that precautions were observed in obtaining representative soil samples for plating. No consistent trends toward definite increases or decreases in numbers of organisms with time of incubation were observed, as may be noted from the analysis of variance in table 2. The nature of the fluctuations is probably attributable in part to competition between various microbial species, to the fact that certain species are capable of developing rapidly for short periods when an abundance of specific, suitable food is available, and also to inherent faults of the plate method as used to determine microbial numbers in soil.

The influence of inherent soil characteristics on microbial activity as manifested by numbers in the specific groups of microbes is convincingly demonstrated in figure 1. The average numbers of bacteria range from 8,825,000 to 325,000 per gram of soil in the A horizons and from 7,650,000 to 453,000 in the B horizons, indicating that certain genetic soil types may be able to support more than 19 times as many bacteria as certain other soil types. The extreme differences in numbers of fungi and actinomyces are still greater than those of the bacteria. The average numbers of fungi vary from 738,750 to 2,133 in the A horizons and from 258,750 to 1,313 in the B horizons, or more than 200 times as many fungi in the soil with the highest count as in the soil with the lowest count. The averages for actinomyces range from 3,800,000 to 4,500 in the A horizons as compared with 1,275,000 to 12,660 in the B horizon, or a ratio of more than 800 to 1 for extremes in the A horizons and about 100 to 1 for extremes in the B horizons.

TABLE 1
Microbial numbers in different climatic soil types
 Numbers per gram oven-dry soil, all decimal fractions dropped

SOIL TYPE	SOIL NUMBER AND HORIZON	DEPTH <i>inches</i>	FUNGI IN THOUSANDS				BACTERIA IN MILLIONS				ACTINOMYCES IN THOUSANDS				CELLULOSE-DECOM- POSING BACTERIA			
			3/6	3/30	4/27	5/25	3/6	3/30	4/27	5/25	3/6	3/30	4/27	5/25	3/6	3/30	4/27	5/25
Ephrata sandy loam.....	1 A	0 -10	39	50	20	22	7	3	3	3	3250	2600	1300	1300	475	135	480	1145
	1 B	10 -36	6	30	10	7	2	3	<1	2	6300	900	400	200	30	175	185	1000
	2 A	0 -15	4	<1	1	2	10	6	3	4	900	900	900	300	5	230	200	75
Mohave clay loam.....	2 A	15 -30	7	4	3	2	3	16	12	6	125	800	1000	300	10	155	225	140
	2 B	40 -50	3	2	2	2	4	15	4	4	205	1000	350	500	20	270	310	265
	3 A	0 -12	30	30	28	31	6	4	2	2	2100	2400	1800	600	85	250	5	300
Ritzville fine sandy loam.....	3 B	12 -24	3	9	9	10	6	10	3	1	490	900	800	400	5	500	525	1025
	4 A	1 -12	18	9	7	7	6	5	4	3	3500	2100	2600	1300	15	205	155	250
	4 B ₁	16 -28	1	<1	1	1	5	6	7	3	1050	1200	1100	500	5	240	40	60
Barnes clay loam.....	4 B ₂	28 -54	1	<1	2	2	4	7	4	5	280	900	800	500	0	20	60	115
	5 A	0 -8	55	50	30	50	6	4	1	3	2500	2700	500	1200	162	145	280	855
	5 B	8 -18	12	10	6	6	9	5	4	2	2000	1700	700	700	50	160	235	510
Garrison gravelly loam.....	6 A	0 -12	60	20	60	3	4	1	4	1	1500	300	1500	800	15	65	55	15
	6 B	18 -40	25	25	20	8	4	1	4	1	700	600	1600	400	25	155	2	300
	7 A	0 -11	51	45	21	11	9	11	11	2	1150	1700	2000	900	125	175	210	200
Houston clay.....	7 B	11 -33	8	7	4	3	9	8	9	3	550	1400	1900	650	55	130	130	150
	8 A	0 -12	165	220	150	180	1	1	<1	<1	380	500	200	20	40	205	280	145
	8 B	12 -24	100	120	90	80	3	1	<1	<1	180	2000	200	20	40	95	225	165
Spanaway gravelly sandy loam*.....	9 A	0 -16	65	210	230	180	<1	<1	<1	<1	400	1750	2000	800	110	275	225	165
	9 B	16 -30	85	30	90	70	2	1	1	<1	120	60	200	400	10	530	200	560
	10 A	0 -8	40	70	20	10	13	4	1	4	900	1600	600	900	45	755	835	230
Spanaway gravelly sandy loam†.....	10 B	8 -28	20	19	20	13	1	5	2	2	310	1400	900	700	125	35	820	800
	10 C	28+	5	7	7	4	<1	1	1	1	7	30	40	35	200	600	25	1265
	11 A	0 -6	90	9	80	80	1	1	<1	<1	490	1300	1000	1100	100	80	140	130
Alderwood loam.....	11 B	6 -20	21	12	20	16	1	<1	<1	<1	85	70	80	230	110	100	170	290
	12 A	0 -10	785	690	690	790	7	3	4	3	4900	4400	3600	2300	70	125	100	125
	12 B	15 -35	325	260	290	160	1	<1	<1	<1	1400	800	300	370	100	115	50	105
Rainier sandy loam.....	13 A	1.5 -5	175	160	180	170	<1	<1	<1	<1	400	60	35	10	500	1200	1045	
	13 B	8 -18	415	130	70	40	<1	<1	<1	<1	5	70	30	40	20	650	70	1600
	14 A ₁	0 -3	85	160	1	160	<1	<1	<1	<1	8	3	2	3	40	35	3	25
Lakewood sandy loam.....	14 A ₂	3 -17	400	460	210	220	<1	<1	<1	<1	3	25	10	55	120	30	130	
	14 B	17 -33	13	20	22	16	<1	<1	<1	<1	20	3400	2000	2400	40	190	280	175
	14 C	31+	14	25	4	8	<1	<1	<1	<1	1850	3000	2000	600	20	85	160	110
Greenville fine sandy loam.....	15 A	0 -3	210	150	190	90	6	2	1	1	850	6000	1170	1100	3	125	145	80
	15 A ₂	3 -12	45	47	25	38	10	6	1	1	700	1000	1170	1100	3	125	145	80
	15 B ₂	24+	7	6	6	6	1	5	1	1	700	1000	1170	1100	3	125	145	80

* Prairie soil. † Forest soil.

The analysis of variance results in table 2 show that the differences in numbers of bacteria, fungi, and actinomycetes among soils in the same climatic group and among climatic groups of soils are statistically significant. They

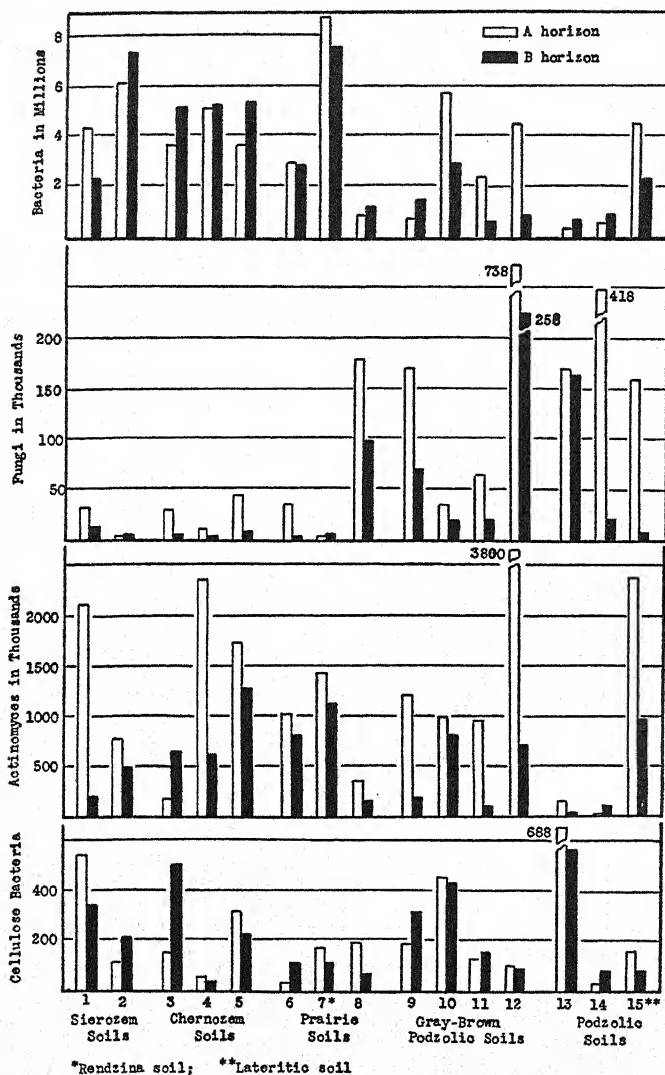


FIG. 1. MICROBIAL NUMBERS PER GRAM OF OVEN-DRY SOIL

Averages of four monthly counts in 2-kgm. quantities of soil maintained at normal moisture capacity at 28°C.

are significant also for the fungi and the actinomycetes between the A and B horizons, but not for the numbers of bacteria between these horizons. The population of aerobic cellulose-decomposing bacteria as determined by the

silica jel plate method was small in all soils, and therefore, the difference in the numbers in the various profiles may not be very significant, although statistically the differences between soils are highly significant, as may be noted in table 2. The large differences in numbers of bacteria, fungi, and actinomyces cannot be attributed to excessive differences in soil reaction, for as shown by the data in table 3, none of the pH values indicates extreme acidity or alkalinity. The values for the oxidation-reduction potentials, which are also recorded in this table, fluctuate considerably but without any definite relation to the large differences in numbers of organisms or to the general character of the broad genetic soil types.

TABLE 2

Results of analysis of variance of microbial numbers in different climatic soil types

ORGANISM	SOURCE OF VARIATION	DEGREE OF SIGNIFICANCE
Bacteria.....	Within dates (error)	..
	Between soils	**
	Between horizons	..
	Between climatic soil groups	**
Fungi.....	Within dates (error)	..
	Between soils	**
	Between horizons	**
	Between climatic soil groups	**
Actinomyces.....	Within dates (error)	..
	Between soils	**
	Between horizons	**
	Between climatic soil groups	*
Cellulose-decomposing bacteria.....	Within dates (error)	..
	Between soils	**
	Between horizons	..
	Between climatic soil groups	..

* Significant.

** Highly significant.

Although there was a marked difference in numbers composing the various microbial groups in different soil types within any single broad genetic group, figure 1 discloses that in general the bacterial population in the solum of the sierozem and grassland soils was much greater than that in the forest soils, and that the reverse was true with respect to the fungi. This is in conformity with the observations of many investigators, except that it does not apply strictly to soil 8, the Spanaway prairie soil, which is atypical in this respect as well as in certain other respects, and behaved more like a forest soil in its influence on microbial activity. The actinomyces were generally more numerous in the grassland soils than in the sierozems or forest soils. The

influence of genetic soil types on numbers of actinomycetes, though significant, was not nearly so pronounced as it was on bacterial and fungal activity, except

TABLE 3
Ammonia nitrogen, nitrate nitrogen, and Eh and pH values of soils at different periods of the experiment

SOIL TYPE	SOIL NUMBER AND HORIZON	DEPTH	NH ₃ -N		NO ₃ -N		Eh(pH ₂)* BEGIN- NING	pH BEGIN- NING
			Begin- ning	End	Begin- ning	End		
		<i>inches</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>		
Ephrata sandy loam	1 A	0 -10	12.5	6.0	2.9	27.6	495.1	7.1
	1 B	10 -36	5.4	6.0	1.2	6.2	497.2	7.0
	2 A	0 -15	10.0	6.8	7.6	8.9	525.5	7.9
Mohave clay loam	2 A & B	15 -30	8.3	5.6	20.8	12.5	560.2	7.5
	2 B	40 -50	11.5	7.1	13.1	8.6	477.4	7.9
Ritzville fine sandy loam	3 A	0 -12	13.6	15.0	1.2	38.1	483.0	6.6
	3 B	12 -24	10.7	13.6	0.7	12.5	547.8	7.2
	4 A	1 -12	10.0	10.0	2.7	85.7	547.8	6.9
Barnes clay loam	4 B ₁	16 -28	8.3	6.8	0.7	11.8	544.4	7.9
	4 B ₂	28 -54	8.8	6.8	0.8	5.3	599.4	7.6
Garrison gravelly loam	5 A	0 - 8	14.3	4.1	2.4	66.6	487.8	7.5
	5 B	8 -18	10.7	4.3	1.4	20.0	522.2	7.0
Palouse silt loam	6 A	0 -12	3.0	3.0	0.9	50.0	482.3	6.4
	6 B	18 -40	3.4	3.3	1.1	5.0	598.2	6.6
Houston clay	7 A	0 -11	13.6	8.8	1.1	10.0	567.0	7.4
	7 B	11 -33	12.5	15.5	0.9	6.9	533.2	7.7
Spanaway gravelly sandy loam†	8 A	0 -12	5.2	2.9	1.7	4.4	526.5	5.0
	8 B	12 -24	2.4	2.4	0.6	13.3	476.8	6.0
Spanaway gravelly sandy loam‡	9 A	0 -16	3.0	3.0	1.6	23.5	439.4	5.4
	9 B	16 -30	2.3	3.2	0.8	1.9	425.4	6.7
Lynden sandy loam	10 A	0 - 8	4.2	4.2	1.0	13.2	454.2	6.6
	10 B	8 -28	4.7	4.7	0.6	8.9	457.0	6.6
	10 C	28+	4.4	4.0	0.4	1.2	424.2	6.7
Alderwood loam	11 A	0 - 6	4.5	2.8	0.8	16.7	388.6	5.9
	11 B	6 -20	4.7	2.5	0.7	1.5	378.9	5.9
Helmer silt loam	12 A	0 -10	10.0	4.7	1.1	27.3	440.4	6.2
	12 B	15 -35	10.0	4.4	0.7	6.0	481.6	5.7
Rainier sandy loam	13 A	1.5- 5	5.4	5.1	1.3	0.7	349.9	6.4
	13 B	8 -18	4.4	2.8	0.6	0.8	480.2	6.3
	14 A ₁	0 - 3	8.3	9.4	0.9	2.2	403.8	5.4
Lakewood sandy loam	14 A ₂	3 -17	6.5	8.3	0.9	0.5	438.4	6.4
	14 B	17 -33	7.1	6.0	0.4	3.2	432.4	6.2
	14 C	31+	7.5	7.1	0.2	1.7	429.6	6.5
Greenville fine sandy loam	15 A	0 - 3	7.9	8.3	1.0	4.8	526.4	6.3
	15 A ₂	3 -12	7.1	8.8	0.4	10.7	565.8	6.4
	15 B ₂	24+	12.5	5.6	0.5	1.6	507.6	5.7

* Calculated at pH value of 2.

† Prairie soil.

‡ Forest soil.

in the two podzol soils. Both of these soils, which are more acid in reaction than the others, contained small numbers.

Another significant point revealed by the graphs in figure 1 is that with proper aeration under suitable moisture and temperature conditions, the B horizons of many of the soils are capable of supporting a very active bacterial flora. This conforms generally with the observations of Brown and Benton (1) and Timonin (9), who found that the B horizon in certain soil profiles contains a larger microbial population than the A horizon. Four of the pedocals and four of the pedalfers, or the majority of the soils used in this study, supported a larger bacterial population in the samples of the B horizon than in those of the A horizon. Both the fungal and actinomyces populations, however, were represented by smaller numbers in most cases in the B horizons than in the A horizons. The magnitude of this reduction in numbers, from the standpoint of all the soils under consideration, is highly significant, as has already been noted. It seems likely also that the material in the C horizon of the soil profiles is much less conducive to microbial growth than the material in the solum. The microbial counts of the two C horizons included in this study strongly indicate this tendency.

*Nature and quantity of humus in relation to genetic soil types and
microbial activity*

The data on the amounts and composition of humus as expressed in terms of carbon and nitrogen are recorded in tables 4 and 5. A comparison of the figures on total organic carbon with the average numbers of microbes as presented in table 1 and figure 1 offers convincing evidence that there is no significant relationship between microbial numbers and humus content of soils belonging to different zonal types or even of different soil types within the same genetic zonal groups. It is apparent that the nature of the humus has a more pronounced effect than the quantity on prevailing microbial species and numbers.

A general concept of the composition and nature of the humus may be gained by a consideration of the carbon-nitrogen ratios in table 6. It is noted that the carbon-nitrogen ratios of the humus substances in the sierozems and grassland soils are distinctly narrower than those of the humus substances in the forest soils, particularly the podzols. This corresponds broadly with the generally more intensive nitrification, as indicated in table 3, as well as with the larger numbers of bacteria and much smaller numbers of fungi in the sierozems and grassland soils, but it does not account either for the generally large differences in numbers of actinomyces between the A and B horizons and among the various genetic soil types or for the small numbers of bacteria and large numbers of fungi in the Spanaway prairie soil, which contains a large amount of humus with a relatively narrow carbon-nitrogen ratio. A better understanding of the chemical composition of the humus in these soils is necessary to explain these differences. This was attempted.

The percentages of total carbon and nitrogen in the various genetic soil types and their different horizons as well as the corresponding amounts of carbon and nitrogen extracted by water, hydrochloric acid, and sulfuric acid solutions

vary greatly, as may be noted from the data in tables 4 and 5. The extremes in total carbon in the A horizons of the various soil types, for example, are 8.9 per cent and 0.75 per cent. The extremes in sulfuric acid soluble carbon are 1.97 per cent and 0.15 per cent. The differences in values for the total and

TABLE 4
Organic carbon fractions in milligrams per gram of oven-dried soil

SOIL NUMBER AND HORIZON	TOTAL ORGANIC CARBON	H ₂ O-SOLUBLE C	HCl-SOLUBLE C	H ₂ SO ₄ -SOLUBLE C	H ₂ SO ₄ -INSOLUBLE C	SUM OF C FRACTIONS	PER CENT RECOVERY
1 A	9.8	0.46	1.76	2.33	5.46	10.0	101
1 B	8.7	0.21	1.12	2.38	5.00	8.6	98
2 A	7.5	0.14	1.01	1.48	4.47	7.1	94
2 A & B	3.9	0.07	0.51	1.00	2.14	3.7	94
2 B	2.9	0.02	0.63	0.46	2.02	3.1	105
3 A	21.9	0.66	5.55	3.78	11.31	21.3	97
3 B	8.2	0.15	2.12	1.43	4.30	8.0	97
4 A	45.5	1.04	10.41	6.90	24.67	43.0	94
4 B ₁	13.9	0.53	2.98	2.59	7.56	13.6	98
4 B ₂	8.2	0.35	1.15	1.99	4.69	8.1	99
5 A	39.0	0.70	5.51	4.18	26.11	36.5	93
5 B	12.0	0.42	2.99	2.86	5.66	11.9	99
6 A	27.9	0.98	5.51	9.21	11.32	27.0	96
6 B	10.8	0.40	2.19	1.51	6.22	10.3	95
7 A	20.1	0.72	4.23	2.16	12.79	19.9	99
7 B	13.8	0.32	3.97	1.28	7.93	13.5	97
8 A	85.9	0.73	12.62	19.73	46.61	79.7	92
8 B	30.3	0.40	9.17	9.35	10.81	29.7	98
9 A	89.7	0.68	20.05	9.69	55.97	86.4	96
9 B	7.4	0.39	1.81	1.66	3.38	7.2	97
10 A	13.4	0.44	4.01	2.42	6.39	13.2	98
10 B	12.8	0.57	3.85	2.57	5.11	12.1	94
11 A	20.5	0.35	5.99	4.05	8.78	19.1	93
11 B	16.1	0.38	4.81	3.54	6.79	15.5	96
12 A	17.3	0.77	3.39	2.78	10.24	17.2	99
12 B	7.4	0.26	0.88	1.59	4.35	7.0	94
13 A	14.1	0.60	1.82	2.61	9.03	14.0	99
13 B	7.3	0.13	1.58	0.87	4.45	7.0	96
14 A ₁	12.3	0.49	1.73	2.35	7.23	11.8	95
14 B	5.6	0.05	1.82	1.19	2.74	5.8	102
14 C	4.0	0.09	0.49	1.06	2.51	4.1	103
15 A	17.2	0.46	2.95	2.67	10.97	17.0	98
15 A ₂	7.6	0.13	1.01	2.09	4.38	7.6	99

sulfuric acid soluble nitrogen are still larger than those of the corresponding values for carbon. Similar variations occur in the values for total carbon and nitrogen and for carbon and nitrogen in the hydrochloric and sulfuric acid extracts of the other horizons. The differences in the notably small quantities of carbon and nitrogen in the hot-water extracts are not so pronounced and

are, therefore, less important. Despite the fact that the variations in amounts of total carbon and nitrogen and of carbon and nitrogen in the organic matter fractions of the different genetic soil types and horizons are large, they fail to

TABLE 5
Nitrogen fractions in milligrams per gram of oven-dried soil

SOIL NUMBER AND HORIZON	TOTAL NITROGEN	H ₂ O-SOLU- BLE N	HCl-SOLU- BLE N	H ₂ SO ₄ - SOLUBLE N	H ₂ SO ₄ - INSOLUBLE N	SUM OF N FRACTIONS	PER CENT RECOVERY	PROTEIN N
1 A	0.62	0.019	0.213	0.163	0.255	0.63	103	0.63
1 B	0.40	0.009	0.129	0.124	0.133	0.39	97	0.38
2 A	0.49	0.011	0.100	0.111	0.274	0.49	99	0.48
2 A & B	0.34	0.004	0.067	0.158	0.119	0.34	100	0.34
2 B	0.27	0.003	0.083	0.053	0.135	0.27	101	0.27
3 A	1.47	0.037	0.433	0.432	0.588	1.49	100	1.45
3 B	0.65	0.012	0.152	0.217	0.256	0.63	97	0.62
4 A	3.71	0.067	0.892	1.059	1.575	3.59	96	3.52
4 B ₁	0.99	0.014	0.316	0.225	0.459	1.01	102	1.00
4 B ₂	0.44	0.004	0.062	0.142	0.254	0.46	105	0.45
5 A	2.26	0.047	0.495	0.545	1.061	2.14	94	2.10
5 B	1.09	0.020	0.332	0.361	0.430	1.14	104	1.12
6 A	1.64	0.037	0.421	0.527	0.565	1.55	94	1.51
6 B	0.47	0.011	0.140	0.138	0.190	0.47	101	0.46
7 A	1.89	0.031	0.571	0.430	0.881	1.91	101	1.88
7 B	1.18	0.014	0.351	0.275	0.524	1.16	98	1.14
8 A	6.92	0.025	1.490	2.224	2.864	6.60	95	6.57
8 B	2.20	0.010	0.823	0.866	0.487	2.18	99	2.17
9 A	3.91	0.028	1.384	0.861	1.368	3.64	93	3.61
9 B	0.25	0.007	0.119	0.062	0.054	0.24	96	0.23
10 A	0.54	0.009	0.256	0.121	0.165	0.55	101	0.54
10 B	0.40	0.005	0.180	0.082	0.138	0.40	99	0.40
11 A	0.79	0.006	0.289	0.331	0.192	0.81	103	0.81
11 B	0.58	0.013	0.262	0.153	0.140	0.56	97	0.55
12 A	0.76	0.024	0.219	0.161	0.327	0.73	95	0.70
12 B	0.32	0.008	0.077	0.050	0.193	0.32	101	0.32
13 A	0.30	0.007	0.081	0.020	0.056	0.16	84	0.15
13 B	0.14	0.005	0.075	0.014	0.051	0.14	100	0.13
14 A ₁	0.27	0.009	0.099	0.067	0.087	0.26	97	0.25
14 B	0.09							
14 C	0.09							
15 A	0.71	0.023	0.267	0.167	0.283	0.74	102	0.71
15 A ₂	0.23	0.013	0.108	0.059	0.055	0.23	101	0.22

show any significant relation to the observed differences in kinds and number of microbes of corresponding genetic soil types and horizons.

In contrast to the relatively large differences in total amounts of humified organic substances, their chemical composition in terms of the different carbon and nitrogen fractions calculated in per cent of the total carbon and nitrogen is comparatively uniform. The quantities of water-soluble carbon and

nitrogen in percentages of the totals are uniformly small. There is some fluctuation in the values for sulfuric acid insoluble, sulfuric acid soluble, and hydrochloric acid soluble carbon and nitrogen. The values for the sulfuric acid insoluble fractions in the A horizons range from 63 to 40 per cent for

TABLE 6
Carbon-nitrogen ratios of soil humus and humus fractions

SOIL NUMBER AND HORIZON	C/N RATIO OF				
	Total organic matter	H ₂ O-soluble organic matter	HCl-soluble organic matter	H ₂ SO ₄ -soluble organic matter	H ₂ SO ₄ -insoluble organic matter
1 A	15.8	24.3	8.3	14.3	21.4
1 B	21.6	24.0	8.7	19.3	37.6
2 A	15.1	13.0	10.1	13.4	16.3
2 A & B	11.3	19.0	7.7	6.4	18.0
2 B	11.1	6.7	7.6	8.8	15.0
3 A	14.8	18.0	12.8	8.8	19.2
3 B	12.6	13.0	14.0	6.6	16.8
4 A	12.3	15.6	11.7	6.5	15.7
4 B ₁	14.1	38.1	9.4	11.5	16.5
4 B ₂	18.7	87.8	18.6	14.0	18.5
5 A	17.3	15.0	11.1	7.7	24.6
5 B	11.0	21.4	9.0	7.9	13.2
6 A	17.0	26.5	13.1	17.5	20.0
6 B	23.1	36.5	15.7	11.0	32.8
7 A	10.6	23.3	7.4	5.0	14.5
7 B	11.7	23.0	11.3	4.7	15.1
8 A	12.4	29.2	8.5	8.9	16.3
8 B	13.8	40.9	11.2	10.8	22.2
9 A	22.9	24.4	14.5	11.3	40.9
9 B	29.8	55.7	15.3	26.8	62.7
10 A	24.8	49.6	15.7	20.0	38.7
10 B	31.8	115.2	21.4	31.4	37.1
11 A	25.8	58.8	20.7	12.3	45.8
11 B	27.7	29.2	18.4	23.2	48.6
12 A	22.6	32.3	15.5	17.3	31.3
12 B	23.2	32.8	11.5	31.9	22.5
13 A	47.0	86.4	22.5	130.6	161.3
13 B	50.7	26.0	21.2	62.2	87.4
14 A ₁	45.8	54.8	17.5	35.2	83.1
14 B	63.2				
14 C	42.5				
15 A	24.1	20.4	11.1	16.0	38.8
15 A ₂	32.9	10.3	9.4	35.6	79.7

carbon and from 55 to 25 per cent for nitrogen. The values for the B horizons vary from 67 to 36 per cent for carbon and from 60 to 22 per cent for nitrogen. Variations of similar character appear in the values for carbon and nitrogen in the sulfuric acid soluble and hydrochloric acid soluble fractions. The differences for any genetic group of soils are not outstanding, however.

Waksman and Hutchings (12) found podzol soils to be high in sulfuric acid soluble and hydrochloric acid soluble carbon and low in protein substances, whereas sierozem and chernozem soils were found to be high in sulfuric acid insoluble carbon and in protein nitrogen but low in sulfuric acid soluble and hydrochloric acid soluble carbon. Remezow and Wlasowa (8) obtained a relatively high percentage of protein nitrogen in proportion to the lignin-humus compounds in the organic matter of Russian gray desert soils but not in the chestnut and chernozem soils. They were unable to extract any appreciable amounts of cellulose compounds from the gray desert and chestnut soils and detected no significant amounts of hemicellulose compounds in the gray desert soils, although these compounds made up about 34 per cent of the organic matter in the chestnut soils. Presumably, the hemicellulose compounds are those which are soluble in a 2 per cent hydrochloric acid solution, and the cellulose compounds those soluble in a 70 per cent sulfuric acid solution. The foregoing results indicate that the composition of the organic matter in Russian desert and semidesert types of soils differs from that of the organic matter in similar American soils, which apparently are not alike in different parts of the United States. The Ephrata and Mohave series, which are classified as gray desert soils, and the Ritzville series, which resembles the chestnut type of soil, contain appreciable quantities of hydrochloric acid soluble and sulfuric acid soluble organic matter, and the proportion of protein nitrogen to lignin-humus compounds in the two sierozem soils does not differ significantly from that in the chestnut and chernozem types of soil, as may be noted in tables 4 and 5 and figures 2 and 3.

The combined values for sulfuric acid soluble and hydrochloric acid soluble nitrogen and carbon appear to be generally larger for the soils in the gray-brown podzolic group than for those of the other genetic groups. It may be noted in this connection that in a previous study (11) the percentages of sulfuric acid soluble carbon and nitrogen, which included hydrochloric acid soluble carbon and nitrogen, varied to a much greater extent in the organic residues of different soils than in the humified organic matter of the homologous soils, and the organic residues containing the largest percentages of sulfuric acid soluble carbon and nitrogen appeared to favor the growth of actinomyces. Evidently the relatively small differences in the percentages of carbon and nitrogen in the sulfuric acid and other extracts as revealed by differential analysis of the humified organic matter in the various genetic soil types and horizons used here are inadequate to explain the observed differences in the nature and activity of the soil microflora.

The available data on the composition of soil organic matter reveal marked differences in the carbon and nitrogen contents of extracts obtained from similar genetic soil types in different parts of the world when the same reagent, such as 2 per cent hydrochloric acid or 70 per cent sulfuric acid, and the same analytical procedures are used. Moreover, the extracts obtained from the organic matter of different genetic soil groups, or from similar soil types developed in separate

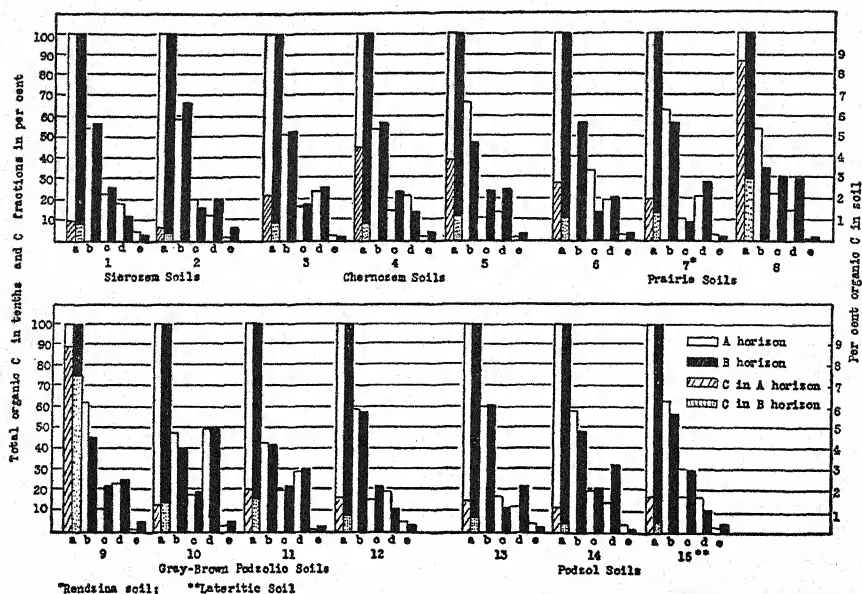


FIG. 2. ORGANIC CARBON FRACTIONS IN PER CENT OF THE TOTAL ORGANIC CARBON
a, total C; b, H_2SO_4 -insoluble C; c, H_2SO_4 -soluble C; d, HCl-soluble C; and e, hot water soluble C.

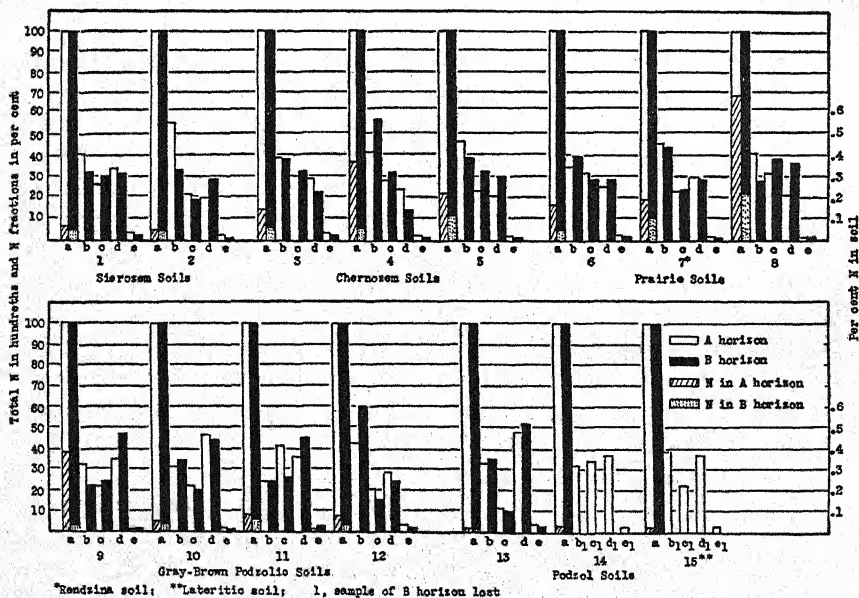


FIG. 3. NITROGEN FRACTIONS IN PER CENT OF THE TOTAL NITROGEN
a, total N; b, H_2SO_4 -insoluble N; c, H_2SO_4 -soluble N; d, HCl-soluble N; and e, hot water soluble N.

areas, by any of the commonly employed specific reagents may yield compounds which are dissimilar in chemical composition. It may be observed from the data in table 6 that the carbon-nitrogen relationships in the various extracts from the soils in any of the specific genetic groups do not vary in the same proportion. If the soils differ in composition with respect to the carbon and nitrogen compounds, probably they differ also with respect to the composition of other compounds which are vital as microbial food. The fact that in general the carbon-nitrogen ratios of the total, hot water soluble, hydrochloric acid soluble, sulfuric acid soluble, and sulfuric acid insoluble organic matter are much greater for the gray-brown podzolic and podzol soils than for the sierozem and grassland soils, and correspond generally with much smaller bacterial populations and much larger fungal populations in the forest soils than in the grassland soils, may indicate that these highly carbonaceous substances are more readily attacked by the fungi than by the other groups of soil microbes. It does not account for the other marked variations in microbial numbers.

Like amounts of carbon and nitrogen in extracts obtained from different soils by any of the specific reagents used in soil organic matter fractionation do not prove that the microbial food value of the substances in the two extracts is identical. Nitrogen and carbon are only two of a number of food elements needed for microbial development. Identical quantities of carbon and nitrogen, which presumably might be found in the hydrochloric acid or any other extracts from two different soils, would not necessarily indicate that these elements are present in the same chemical combinations or are equally well balanced with other essential food elements in corresponding extracts from the two soils. Obviously, the soil that would yield extracts in which the carbon and nitrogen compounds are in forms especially suitable for microbial food and properly balanced with the other essential nutrients in the extracts, would be much more suitable for microbial development than the one yielding corresponding extracts containing carbon and nitrogen compounds identical in quantity with those in the first, but not so suitable for microbial food and not properly balanced with the other essential nutrients in the extracts. The fractionation method of soil organic matter analysis is not sensitive enough to detect these differences and, therefore, to account for certain important differences in the composition of the soil microflora and in the activity of specific groups of soil microbes. Unquestionably, differences in the nature of soil humus occur, and these differences may exert a marked and significant influence on microbial activities, inherent soil properties, and soil fertility, but the nature of the specific qualities of the humus causing these effects, though of primary importance and certainly worthy of further study by improved methods of soil organic matter analysis, is still obscure.

SUMMARY

Microbial and chemical determinations were made on representative samples of various horizons of 15 soil types to study the numbers of organisms in the

principal microbial groups in relation to the nature and chemical composition of the humified organic matter.

The influence of inherent soil properties on microbial activity was expressed by large differences in numbers of bacteria, fungi, and actinomyces in the A and B horizons. Certain genetic soil types supported 19 times as many bacteria, 200 times as many fungi, and 800 times as many actinomyces as certain other types. The numbers of aerobic cellulose-decomposing bacteria were small in all soils, and only four soils contained active *Azotobacter*.

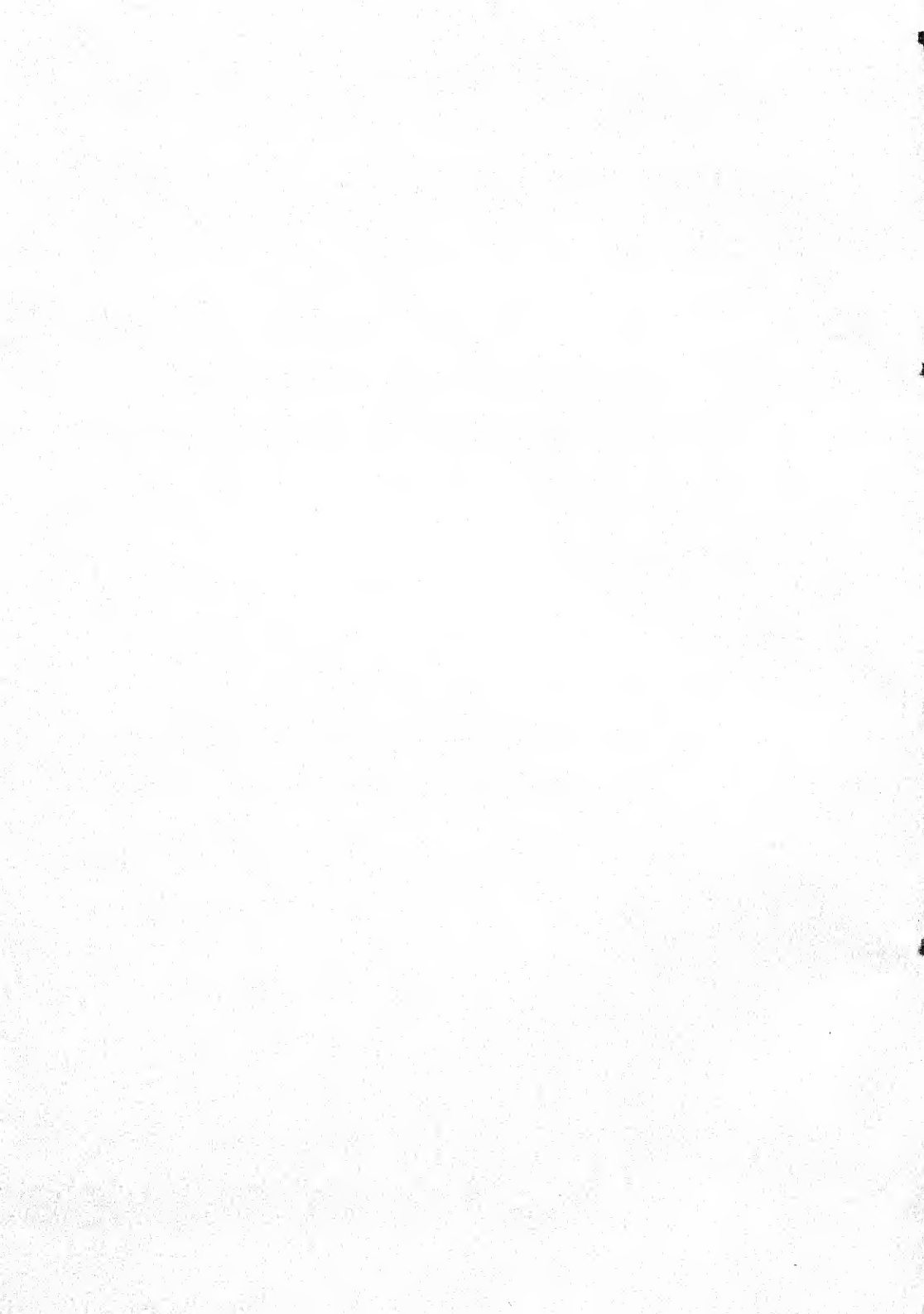
The bacterial population was generally larger and the fungal population generally much smaller in the solum of the sierozem and grassland soils than in the solum of the forest soils, particularly the podzols. The larger bacterial population corresponded broadly with the greater rate of nitrification in, and the narrower carbon-nitrogen ratios of the humified organic matter and organic matter fractions of, the grassland soils. Under the conditions of the experiment, in which aeration, soil moisture, and temperature were properly controlled, four of the pedocals and four of the pedalfers, or the majority of the soils used in this study, supported a larger microbial population in the B than in the A horizon.

No significant relation existed between the total content of humified organic matter in the soils and their microbial numbers. Although differences in composition of the humus in the various genetic soil types and horizons were revealed by fractionation of the humified organic matter and these differences corresponded broadly with the activity of certain microbial groups, the method of fractionation used was inadequate to detect those more delicate differences in the nature and composition of humus that are responsible for many of the observed differences in microbial numbers in various genetic soil types and in various soil types within the same genetic soil groups.

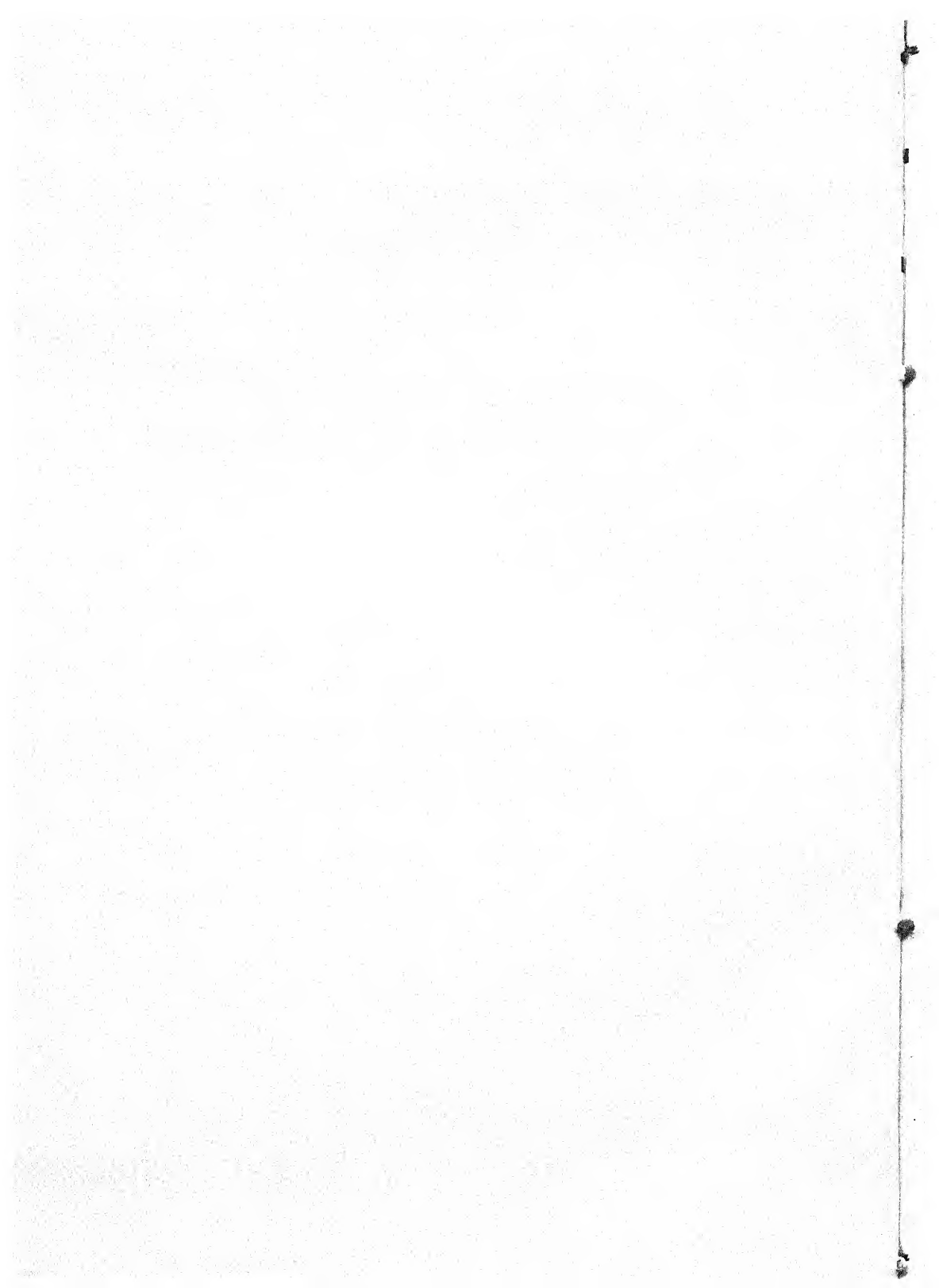
REFERENCES

- (1) BROWN, P. E., AND BENTON, T. H. 1930 Microbial studies of some typical Iowa soil profiles. *Iowa Agr. Exp. Sta. Res. Bul.* 132: 363-420.
- (2) JOFFE, J. S., AND WATSON, C. W. 1933 Soil profile studies: V. Mature podzols. *Soil Sci.* 35: 313-331.
- (3) MAL'CHEVKAIA, N. I. 1935 Microbiological characters of certain types of forest soils. *Pedology* [Russian] 3: 225-239. [*Biol. Abs.* 9: 5355 (1935).]
- (4) MARBUT, C. F. 1935 Atlas of American Agriculture: Part III. Soils of the United States. U. S. Department of Agriculture.
- (5) NIKIFOROFF, C. C. 1937 The inversion of the great soil zones in western Washington. *Geogr. Rev.* 27: 200-213.
- (6) RAZUMOV, A. S., AND REMEZOV, N. P. 1928 Distribution of microorganisms in the profiles of podzol soil. *Jour. All Russ. Cong. Bot.* 1928: 210. [*Biol. Abs.* 5: 14840 (1931).]
- (7) REMEZOV, N. P. 1933 Beiträge zur Charakteristik der organischen Substanz der Böden der Ud SSR. I. N:C Verhältniss in der organischen Substanz der Böden der Ud SSR. *Ztschr. Pflanzenernähr., Düngung, u. Bodenk.* (A) 30: 285-299.

- (8) REMEZOW, N. P., AND WLASOWA, M. M. 1933 Beitrage zur Charakteristik der organischen Substanz der Böden der Ud SSR. II. Annähernde qualitative Zusammensetzung der organischen Substanz der Hauptbodenarten der Ud SSR. *Ztschr. Pflanzenernähr Düngung, u. Bbenk.* (A) 30: 299-311.
- (9) TIMONIN, M. I. 1935 The microorganisms in profiles of certain virgin soils in Manitoba. *Canad. Jour. Res., Sec. Bot. Sci.* 13: 32-46.
- (10) VANDECAVEYE, S. C., AND BAKER, G. O. 1938 Microbial activities in soil: III. Activity of specific groups of microbes in different soils. *Soil Sci.* 45: 315-333.
- (11) VANDECAVEYE, S. C., AND KATZNELSON, H. 1938 Microbial activity in soil: IV, V. *Soil Sci.* 46: 57-74; 46: 139-167.
- (12) WAKSMAN, S. A., AND HUTCHINGS, I. J. 1935 Chemical nature of organic matter in different soil types. *Soil Sci.* 40: 347-363.
- (13) WAKSMAN, S. A., AND STEVENS, K. R. 1930 A system of proximate chemical analysis of plant materials. *Indus. and Engin. Chem.* 2: 167.
- (14) WILDERMUTH, R., PASCO, R. E., AND HUBBARD, E. H. 1939 Soil Survey of Kitscap County, Washington. *U. S. Dept. Agr. Soil Survey Ser.* 1934, No. 12: 1-41.







CONCRETIONS AND REFRACTORY DEPOSITS IN SOME NATAL COASTAL SOILS

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Concretions of some kind are found in most soils, where they have been formed by secondary processes. The movement of water, carrying with it the soluble soil constituents, mostly iron, and the subsequent precipitation of these solutes at lower (or higher) levels due to new chemical and physical properties arising in the soil profile, are generally responsible for the presence of these very characteristic formations.

Concretions are very common in the Natal and Zululand coastal soils, but hitherto no study of them and no attempt to classify them appear to have been made. Before such a classification is attempted something must first be said about the soil types in which these concretions occur and also about the environmental conditions under which these soil types have been formed. A possible relationship between soil concretions and soil types can then be more intelligibly discussed.

Soil concretions cannot be dissociated from the genetic soil profile; hence in classifying them it is essential to describe the environment in which they have been built up. This environment comprises the parent material, which has played its part and is now largely out of the picture, and the varied pedogenic processes influenced principally by climate. The geological factor, however, is not entirely dismissed, and is in fact preferred by the writer as the basis of soil classification in the perhaps limited sphere of the 300-mile coastline of the sugar belt of Natal and Zululand. Here, where most of the soils are primary, there is such a close association between the parent rock and its soil that it is generally possible to identify soil types and name their geological origin after a brief field inspection. Along the sugar belt the climate, furthermore, is uniform, yet very distinct primary soil types exist; to account for them entirely on a climatic basis, therefore, is difficult. The modifying effect of parent material exceeds other factors in importance and thus leads to the possible identification of soil concretions with certain geological soil types.

The sphere of pedogenic processes is the soil profile, and it is here that we can imagine a continuous age-old transformation taking place. Rainfall, temperature, and evaporation, among other agencies, transform an otherwise homogeneous parent material into those characteristic layers or horizons which

¹ Publication authorized by the director.

make up the soil profile. Because of the hilly nature of the coastal belt, this profile is generally shallow, and soils of the deep phase are met with mostly in intrusive areas and more particularly in the alluvial deposits along river valleys near the sea. No concretions have been encountered in secondary soils; hence the probability of an association between concretions and geological formations is strengthened.

Weather observations at this Experiment Station and at other points on the coast² indicate that the mean annual rainfall is 41.9 inches, the majority of which falls during the warm summer months from November to March inclusive. The mean maximum temperature varies from 81.2°F. in February to 71.5° in July, and the mean minimum temperature ranges from 66.7° in February to 51.7° in July. The average annual temperature is 68.6°, ranging from 75.0° in February to 60.0° in July. The mean temperature of the earth was found to be 71.6° at 1 foot (range 78.4° in March to 63.1° in July), 72.6° at 2 feet (range 80.0° in January to 65.2° in July), and 72.9° at 4 feet (range 78.5° in March to 66.8° in August). The normal annual humidity is 73.6 per cent at 8.30 a.m. and 63.0 per cent at 1 p.m. Evaporation exceeds rainfall by several inches.

In the geological classification of the Natal and Zululand coastal soils the following formations are recognized, lying, with the exception of the Ecce series and the Dwyka conglomerates, uncomformably in order of increasing depth: Ecce shales, Dwyka conglomerates, Table Mountain sandstone, granite, and basalt. The last is intrusive in all formations, but more particularly so in the Ecce series. Overlying the original geological formations along the entire coastline and extending inward to distances varying from a few hundred yards to five or six miles is a deposit of considerable depth and of uncertain origin, geologically known as Wind-Blown or Recent sands. This soil type, it must be pointed out, cannot be associated with any parent rock, but it is very characteristic and very readily distinguished from all other soil types by its uniformly red color and by such other characteristics as will be discussed later. In the northern extremity of the sugar belt, where there are extensive low-lying areas, the soil type becomes a rather unproductive light gray sand. Though the Recent sands may not be strictly primary, they are regarded as such, since their origin is not altogether clear.

Though concretions occur very widely in all types of soils under certain conditions, the writer has formed the conclusion that their existence in any particular region can be traced to, and associated with, geological soil types. In accordance with this view the principal types and subtypes of concretions will be described and their association with soil types discussed. Table 1 summarizes the observations made.

All mechanical analyses of soil profiles reported in this paper were made by

* Beater, B. E. 1939 Summary of weather conditions over 11 years (1928-38) in the sugar belt of Natal and Zululand. *Proc. So. African Sugar Tech. Assoc.* 1939: 64-71.

TABLE 1
Concretions and related soil types

CONCRETION	POSITION IN PROFILE	SOIL	GENERAL MORPHOLOGY OF DESCENDING PROFILE					Moisture
			Color	Consistence	Depth	Texture	Structure	
Ironstone gravel	Compact or loose layer below A horizon	Ecce series*	Gray into yellow	Compact and cemented into plastic and sticky	Shallow*	Sandy into silty clay	Pulverized into cloddy	Slightly moist
Rubble	Below A horizon	Dwyka conglomerate*	Gray into yellow	Compact and cemented	Shallow	Sandy into clay loam	Pulverized into cloddy	Moderately dry
Ironstone concretions	Throughout profile	Doleritic and basaltic	Dark brown or red into red*	Compact	Deep	Clay loam into clay	Crumbly and cloddy into nutlike and columnar	Moist or slightly moist
Concretions in Recent sands	Irregular broken layer at any depth	Recent sands	Reddish brown into red	Loose or compact	Deep	Sand or loam into sand or clay loam	Structureless throughout or cloddy throughout	Slightly moist
Bog iron ore	Near water table	Recent sands*	Gray into mottled yellow-gray	Loose	Deep	Sand into sandy clay	Structureless throughout	Wet
Lime concretions	In B horizon	Ecce series*	Dark gray into black	Compact and sticky	Moderately deep	Clay loam into clay	Cloddy and columnar	Moist

* Generally.

means of the pipette method, as modified by the writer;³ and all chemical analyses, by sodium carbonate fusion.

TYPES OF CONCRETIONS

Type 1a. The most common form of concretion is found generally in the soils of the Eccla series. It occurs, usually, in a semicompact layer immediately between the A and B horizons and lying on either rather impervious clay or silty clay. Plate 1, figure 1, illustrates this type of concretion. The gravel layer is clearly visible in the profile. This layer varies in thickness from two to several inches and, except where erosion has taken place, is usually at a fairly constant depth below the surface. In many areas, however, the surface soil has eroded and left the concretions lying over the surface. These loose concretions, variously referred to as ironstone gravel or pistolithic gravel, appear to be similar to the perdigón encountered by Bennett and Allison in Cuba. Furthermore, in some places a double layer has been observed, one layer running parallel a foot or so below the other, particularly where the soil profile is deep. The second layer is less regular, but appears to be similar in all other respects.

The presence of these concretionary layers has not been explained satisfactorily. It is maintained that under favorable climatic conditions water movements lead to the migration and precipitation of elements (chiefly iron) in those layers in which they are found to occur. A puzzling feature, however, is that the concretionary layers are mixed with numerous quartz fragments and crystals, miscellaneous stone fragments, and even water-worn stones, none of which are found outside the concretionary layers. Are chemical agencies thus solely responsible for the presence of ironstone gravel layers?

Under certain conditions the concretionary layer becomes cemented and forms a very hard rocky layer, generally known as hardpan, and in South Africa as "ouklip." This undoubtedly is similar to the macarrero of Bennett and Allison. The cementing material is either iron or humus, in the latter case the formation remaining softer. Where the ouklip lies on the surface through erosion, the agricultural value of the land is considerably impaired.

The pebbles are hard and brittle but pulverize very readily. Some of them are magnetic, and others have only a magnetic core; but the majority are not magnetic at all. Heating does not influence their magnetism. They are highly polished and smooth and vary in color from grayish yellow to reddish brown. Occasionally larger concretions are found, hollow in the center, with a hard outer shell. The concretion illustrated (pl. 2, no. 8) was found, when broken open, to contain a ball of red sandy soil and some round ironstone gravel. Table 9 shows the concretions of type 1a to be composed almost entirely of iron. The absence of manganese is interesting and confirms the work of Prescott.⁴ The nonsilica constituents of the concretions are completely

³ Beater, B. E. 1937 Improved technique in grading of coarse and fine sands during mechanical analysis of soils. *Jour. Agr. Sci.* 27: 123-125.

⁴ Prescott, J. A. 1934 The composition of some ironstone gravels from Australian soils. *Trans. Roy. Soc. So. Aust.* 58: 10-13.

soluble in boiling hydrochloric acid, the results for iron and alumina soluble in hydrochloric acid having been found to be identical with those obtained by sodium carbonate fusion.

The mechanical analysis of a typical profile in which the concretionary layer is encountered is given in table 2.

Type 1b. A subtype in a locality some 60 miles from type 1a was observed. The layer was very loose, was embedded in sandy loam, and could be taken from the soil profile in handfuls merely by scraping with the fingers. These concretions were less regular in shape than those of type 1a, a large proportion being fragmentary. They were dark reddish brown and highly polished. Chemical analysis (table 9) shows them to be lower in iron content than type 1a and also deficient in manganese.

Type 2. The second type of concretion occurs most frequently on dry hilly soils derived from Dwyka conglomerates. In contrast with the first type,

TABLE 2
Mechanical composition of soil profile containing layer of ironstone gravel, type 1a

DEPTH.....inches	0-6	6-12	12-14	14-26
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Coarse sand and gravel.....	13.6	12.4	82.2*	2.1
Fine sand.....	53.4	44.0	4.7	45.8
Silt.....	12.5	11.9	2.2	22.7
Clay.....	15.3	23.3	7.0	21.2
Moisture.....	2.7	5.5	4.1	7.0
Loss by solution (Fe_2O_3 , etc.).....	0.7	0.4	0.2	0.2
Undetermined (organic matter, etc.).....	1.8	2.5	1.0
Total.....	100.0	100.0	100.4	100.0

* Almost entirely concretionary gravel. The pebbles, intimately mixed with the small percentage of clay, form a compact layer.

it forms a less compact layer, which partakes more of the nature of rubble and gives the impression of small rock fragments saturated with iron. The concretions are very irregular, some larger ones being flat and jagged, others thin and pointed. They are yellowish brown externally, but many are reddish brown when broken in two. Their origin is difficult to explain. The layer is well marked and occurs, like the first type, below the A horizon but immediately above the weathering parent material.

The chemical and mechanical analyses of the entire profile in which the rubble layer occurs are shown in tables 3 and 4. That the rubble layer has absorbed iron is indicated in table 3, where the soil layer above contains 3.10 per cent Fe_2O_3 and the layer below 7.55 per cent, as opposed to 20.25 per cent of the rubble layer itself.

Both the ironstone gravel layers and the rubble layers may act as natural underground drains, leaving the upper arable soil very dry. The writer has noticed water running from the gravel layer in a cutting after heavy rains.

A further undesirable feature of these concretionary layers, particularly the first, is that they markedly inhibit root penetration. That phosphorus migrates into these concretions has been shown from an analysis of the surrounding soil. Bennett and Allison in Cuba found in one case ten times more phosphorus in the concretions than in the surrounding soil. Where such migration

TABLE 3
Chemical composition of soil profile containing layer of rubble, type 2

DEPTH.....inches	0-8	8-16	16-20	20-30
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
SiO ₂	76.65	75.45	53.08	57.72
CaO.....	0.52	0.55	0.52	0.50
MgO.....	0.22	0.30	0.40	0.50
K ₂ O.....	2.28	2.26	1.97	1.99
P ₂ O ₅	0.22	0.14	0.23	0.19
TiO ₂	0.39	0.29	0.49	0.60
MnO.....	0.01	Nil	0.05	Trace
Fe ₂ O ₃	2.65	3.10	20.25	7.55
Al ₂ O ₃	8.63	9.27	11.88	17.16
Na ₂ O.....	2.62	2.95	1.82	2.35
Moisture at 105°C...	1.61	1.81	2.47	4.73
Loss on ignition.....	5.16	4.85	6.98	7.24
Total.....	100.96	100.97	100.14	100.53

TABLE 4
Mechanical composition of soil profile containing layer of rubble, type 2

DEPTH.....inches	0-8	8-16	16-20	20-30
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Coarse sand and gravel.....	16.9	17.6	65.1*	10.5
Fine sand.....	48.8	48.6	12.0	27.6
Silt.....	14.2	14.6	11.3	16.9
Clay.....	15.1	16.1	8.4	37.3
Loss by solution (Fe ₂ O ₃ , etc.).....	0.6	0.5	0.2	0.4
Moisture.....	1.6	1.8	2.5	4.7
Undetermined (organic matter, etc.).....	2.8	0.8	0.5	2.6
Total.....	100.0	100.0	100.0	100.0

*Almost entirely rubble.

has taken place the phosphorus is virtually lost to the plant. Experiments on sugar cane at this experiment station have shown no response to phosphatic fertilizers on soils containing the gravel layer of the first type, even though the soil is very low in available phosphorus.

Type 3. Concretions of the third type are scattered through the entire soil profile, being almost absent in the upper section but increasing to a depth

of 5 or more feet. They can be picked out with the fingers, but rarely occur in sufficient quantities to constitute more than a fraction of the soil. They have been noted only in heavy brownish red clayey soils, principally those of doleritic and basaltic origin. They are small, round or irregular and polished, and black or brown-black, a large proportion of them also being magnetic. Un-

TABLE 5
Chemical composition of soil profile containing ironstone concretions of type 3

DEPTH.....inches	0-10	10-26	26-54	54-72
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
SiO ₂	63.51	58.52	45.45	48.78
CaO.....	0.25	0.15	0.12	0.10
MgO.....	0.15	0.17	0.15	0.17
K ₂ O.....	1.00	1.06	0.75	0.95
P ₂ O ₅	0.29	0.26	0.21	0.22
TiO ₂	1.02	1.18	0.91	1.24
MnO.....	0.22	0.14	0.10	0.15
Fe ₂ O ₃	9.20	10.40	11.50	12.50
Al ₂ O ₃	11.47	15.62	23.98	23.19
Na ₂ O.....	0.46	0.34	0.27	0.43
Moisture at 105°C....	4.37	5.06	6.82	4.68
Loss on ignition.....	8.16	8.06	9.51	8.92
Total.....	100.10	100.96	99.77	101.33

TABLE 6
Mechanical composition of soil profile containing ironstone concretions of type 3

DEPTH.....inches	0-10	10-26	26-54	54-72
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Coarse sand and gravel.....	18.9	18.9	7.5	9.0
Fine sand.....	23.8	17.8	24.8	22.2
Silt.....	9.1	7.6	15.5	15.1
Clay.....	38.9	47.3	42.4	47.1
Loss by solution (Fe ₂ O ₃ , etc.).....	0.6	0.3	0.2	0.2
Moisture.....	4.4	5.1	6.8	4.7
Undetermined (organic matter, etc.).....	4.3	3.0	2.8	1.7
Total.....	100.0	100.0	100.0	100.0

doubtedly, these concretions have been formed *in situ* by leaching and precipitation, chiefly of iron. They have never been noted in an initial and possibly soft stage, but always highly polished and hard, giving the impression of a process that has long since ceased.

Tables 5 and 6 show the chemical and mechanical analyses of a typical heavy red basaltic soil profile in which these pebbles were found scattered.

The concretions of type 3 chemically analyzed (table 9) were taken from a

heavy red clayey soil similar to that of tables 5 and 6, where they were scattered between the second and fourth foot of the solum. In chemical composition they resemble very closely the ironstone gravel of the first type. Many of the concretions of the first and third types show concentric fracture on breaking.

Type 4a. The fourth type of concretion occurs widely in profiles of the Wind-Blown coastal sandy soils or Recent sands. This red soil type usually is loose in consistence, becoming more compact with depth.

Table 7 shows the mechanical composition of the two typical phases of this soil type, the compact and the loose. No concretionary layer has been found in the loose phase, though it may exist in some form at greater depths than those reached during soil survey operations.

TABLE 7

Mechanical composition of compact and loose phases of Wind-Blown coastal sandy soils

PHASE.....	COMPACT	LOOSE	COMPACT	LOOSE	COMPACT	COMPACT
Depth.....inches	0-12	0-12	12-42	12-48	42-56	56-72
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Coarse sand and gravel	67.3	61.9	55.7	62.7	39.0	44.2
Fine sand	25.8	27.5	31.4	29.2	17.8	19.0
Silt	1.6	1.6	2.0	1.7	3.3	2.9
Clay	3.8	4.3	8.0	4.4	37.0	30.8
Loss by solution (Fe ₂ O ₃ , etc.)....	0.1	2.4	0.1	1.4	0.1	0.1
Moisture	0.2	0.4	0.6	0.3	2.0	1.6
Undetermined (organic matter, etc.)	1.2	1.9	2.2	0.3	0.8	1.4
Total	100.0	100.0	100.0	100.0	100.0	100.0

In many areas this soil type may also become very compact immediately below the surface. It becomes a deeper red with increasing depth, though in many instances it remains loose and sandy to a depth of more than 8 feet. In the compact or semicompact profile about 6 to 10 feet from the surface occurred the concretions referred to in this paper. They were found in a thin irregular band, or as isolated bodies maintaining a fairly uniform level from the surface. They are also to be found as thin platelike layers running at various angles to the surface. The concretions are hard or moderately hard and many can be broken with the fingers.

Table 8 gives the chemical analysis of a compact Wind-Blown sand profile, very similar to that from which the concretions analyzed were taken. The chemical analyses of the concretions (table 9) show 44 per cent iron oxide, whereas the maximum concentration of iron in the soil itself (table 8) is only 10.40 per cent, indicating that a decided accumulation of this element does take place in these concretions from the soil, though no ironstone gravel has been encountered in the Wind-Blown sands, however clayey and compact they may become.

TABLE 8

Chemical composition of soil profile containing concretions of type 4a

DEPTH.....inches	0-12	12-42	42-56	56-72
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
SiO ₂	92.34	89.38	70.06	74.98
CaO.....	Trace	Trace	0.12	0.17
MgO.....	0.08	0.08	0.25	0.22
K ₂ O.....	0.48	0.52	0.44	0.42
P ₂ O ₅	0.15	0.16	0.09	0.07
TiO ₂	1.76	1.82	1.72	1.24
MnO.....	0.12	0.11	0.15	0.13
Fe ₂ O ₃	3.30	4.20	10.40	8.70
Al ₂ O ₃	1.07	2.12	8.54	7.66
Na ₂ O.....	0.28	0.27	0.19	0.20
Moisture at 105°C.....	0.22	0.57	1.99	1.58
Loss on ignition.....	0.81	1.12	5.43	4.52
Total.....	100.61	100.35	99.38	99.89

TABLE 9

Chemical composition of concretions and refractory residues

TYPE.....	1a	1b	2	3	4a	4b	5a	5b	5c	5d	5e	6	7
Description.	Ironstone gravel	Ironstone gravel	Rubble	Ironstone concretions	Coarse concretions	Coarse concretions	Black residue	Black residue	Black residue	Black residue	Black residue	Bog iron ore	Lime concretions
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
SiO ₂	8.0	23.4	53.1	15.4	40.0	76.5	11.9	29.4	19.8	23.1	27.0	54.8	22.9
Fe ₂ O ₃	67.9	55.4	19.8	68.8	43.0	5.6	42.6	27.7	6.6	40.7	38.2	28.2	3.7
FeO*.....	0.8	0.6	0.4	0.8	1.0	0.8	21.4	13.2	4.9	3.0	9.5	0.6	Nil
Al ₂ O ₃	4.7	5.2	11.9	4.9	3.4	4.6	1.9	0.4	15.8	1.5	1.6	4.7	0.4
P ₂ O ₅	0.4	0.2	0.2	0.2	0.4	0.2	0.4	0.5	0.6	0.2	0.1	0.6	0.4
TiO ₂	0.5	0.6	0.5	0.9	2.2	1.6	22.9	25.1	42.4	28.8	21.6	1.1	0.7
MnO.....	Nil	Trace	0.1	0.4	0.1	1.4	1.0	0.9	0.5	1.0	0.5	1.0	0.1
Moisture at 105°C.	1.8	1.5	2.5	1.8	2.0	1.1	Nil	Nil	0.1	Nil	0.1	1.0	1.6
Ignition loss (or gain) ...	12.4	10.6	7.0	4.6	8.4	3.0	+2.8	+3.2	+1.1	+2.1	+1.5	6.6	29.8
Sum-mation..	96.5	97.5	95.5	97.8	100.5	94.8	99.3	94.0	89.6	96.2	97.1	98.6	59.6

* Soluble in concentrated hydrochloric acid.

The concretions of the fourth type are usually large, of broken and irregular structure, and are found rather deep in the soil. Plate 1, figure 3, illustrates a band of these concretions (type 4a). They do not appear to lie between any

particular horizons, and their rough structure and irregular occurrence suggest that they are of comparatively recent origin, like the soil type in which they are embedded. Stretching in a thin band in cuttings, they have sometimes been found to attain a length of several inches.

Type 4b. Concretions in this same soil type, semicompact Recent sands, were found in a locality some 30 miles away. These concretions vary considerably in size. The specimens photographed and analyzed were largely fragments. In general appearance they resemble in many respects the 4a concretion to which they are undoubtedly allied, but whereas the 4a had semipolished irregular surfaces, the 4b were softer and when broken revealed black patches and layers, suggesting particles of soil cemented together with iron and manganese.

In the same test pit were encountered large layers of the material several feet across and several inches thick. They had an irregular appearance and seemed to be detached in the soil. They were harder than the concretions analyzed and difficult to penetrate with the pick. This concretionary substance was found in a layer about 6 feet below the surface in the red clay loam to loam of the Wind-Blown sands.

Since the first draft of this paper was written, a further variation of the fourth concretionary type has been found by the writer in the compact Recent sands. There were two concretionary layers, the first consisting of semihard irregular bodies almost an inch in diameter. The second layer, about 36 inches below the first, was solid and cemented but soft enough to be cut through with a pick. Plate 1, figure 2, illustrates this double concretionary layer lying at both extremities of the yardstick.

Type 5. The fifth type of concretion partakes of the nature of a refractory residue homogeneously dispersed through the soil, the concentration in sandy soils increasing gradually with depth. It is a shiny black crystalline-looking substance of the approximate dimensions of very fine sand, and when powdered it takes on a purplish hue. The material is found in heavy doleritic soils and in the Wind-Blown or Recent sands, where it is noticeable in the little dry water channels after rains, lying like black coal dust. It is found on certain parts of the seashore and on the beds of neighboring rivers. The origin of this material is not known, since it may have been formed through secondary deposits or derived from basaltic rocks much older than the Eccas, now covered by the Recent sands in which it is so abundantly found. Fusion analyzes (5a, b, c, d, e, table 9) show that it is composed almost entirely of iron and titanium, a large proportion of the iron being in the reduced state. The substance may be identified with ilmenite or titanite iron ore, which it closely resembles in many of its properties. The first sample, 5a, was taken from the seashore on the Natal-Zululand border and was washed free from chlorides before analysis. The second sample, 5b, was taken on a hilltop about 60 miles south of the first, and 3 miles from the seashore. The sample was collected from the surface of the ground after rain. Analyses show these two

substances to be essentially the same, the proportion of silica being governed by the admixture of sand particles. A similar substance was observed by the writer on the South Pacific coast of Australia, and a sample collected on the Queensland border was analyzed by way of comparison. The analysis, 5c, shows a higher percentage of titanium, though the substance is to all appearances identical. The presence of this residue in the Recent sands raises the titanium content of the soil in some instances to as much as 2 per cent. The occurrence of dark concretionary matter of type 4b offers the possibility that this residual substance (type 5) is derived from such secondary formations. That it should be lying in such abundance throughout the Recent sands, with no noticeable outcrops of its parent source, suggests that it has originated from the weathering of secondary concretionary formations. On the other hand, its high titanium content points to the weathering of some primary material, such as dolerite. An analysis of the substance found in unmixed doleritic soil, 5d, shows it to be very similar in composition to that found in the Recent sands and elsewhere. A further sample, 5e, found in heavy red soil of an intrusive nature at the foot of the Drakensberg Range (near Loskop), 150 miles from the coast, also shows a similar composition.

The first sample of type 5 was found to be strongly magnetic, whereas the other samples contained only a small proportion of magnetic particles.

Type 6. The sixth type is common in low-lying waterlogged areas and is sometimes known as bog iron ore. The sample analyzed (table 9) was found at a depth of 8 feet in a rather waterlogged area of the Recent sands. This soil type on exposure to waterlogging conditions becomes very light gray and may contain over 90 per cent silica, as chemical analyses of neighboring soils of this type show. The soil profile in which these concretions are found is a loose phase of the Recent sands which has obviously become leached down to the water table. In this, the concretions lie in a loose layer and can be removed readily with a spade. They are yellow-gray, revealing dark shiny blue and black patches when broken. The soils in which such concretions are found may be described as podzolic. Pea ore, a combination of iron and manganese, is common in such types. These concretions are perhaps too low in manganese to be termed ferromanganiferous, of the type classified by Winters.⁵ They are, nevertheless, roughly ten times higher in manganese content than the surrounding soil. A high accumulation of phosphorus has also taken place in these concretions.

Type 7. The seventh type of concretion was found in heavy dark clayey soil overlying Ecca shales. These concretions are composed largely of lime, as the silicate and carbonate. The samples analyzed contained 33 per cent CaO. These concretions, or nodules, are loosely embedded in the clay in the horizon immediately overlying the weathering shale. They are brittle and when broken show the structure illustrated by the smaller of the two concre-

⁵ Winters, E. 1938 Ferromanganiferous concretions from some podzolic soils. *Soil Sci.* 46: 33-40.

tions in plate 2, no. 7. The accumulation of calcium in the B horizon, it may be observed, is a characteristic chemical feature of chernozem, or black earth, under conditions of low rainfall.

DISCUSSION

In general, concretions and concretionary materials are scattered throughout soils, as hard or semihard bodies or as soft dark patches, lending a mottled appearance to the profile in which they are found. They are distinguished from primary materials such as stone fragments and pebbles, which are obviously of transported origin.

Three main factors control the formation of concretions; namely, soil type and composition, climate, and topography. These factors have been discussed and amply illustrated in this paper. Distinct forms of concretions are found in each soil type, and so regular is the relationship that it is generally possible with a little experience to predict roughly the geological soil type (irrespective of all other factors) in which the concretions were formed. The classification given here is not intended to be final. Though the wide field of research embodied may stress the incompleteness of any such individual attempt as this, it may also open the way to further and more thorough investigations later. At any rate, it seems desirable to obtain some sort of classification of these very abundant secondary formations, which cannot be disregarded in any systematization of soil groups.

SUMMARY

A study has been made of concretions and residues, principally in Natal and Zululand coastal soils, under the following headings:

- 1 (a, b). Ironstone gravel, found in layers in soils of a podzolic type overlying shales, generally between the A and B horizons.

2. Rubble, found in moderately dry shallow soils between the A and B horizons, overlying Dwyka conglomerates.

3. Ironstone concretions, found sparsely scattered through heavy deep clayey soils of lateritic type.

- 4 (a, b). Coarse concretions in Recent sands, irregularly distributed in compact soil in a thin or scattered layer, or as a broken cemented layer, or as a double concretionary layer.

- 5 (a, b, c, d, e). Black residue found in the entire profile of Recent sands, on neighboring seashores and river beds, in Australia, and in doleritic soils at two distant points in Natal.

6. Bog iron ore, found in layers near the water table in poorly drained Recent sands.

7. Concretions of lime found in heavy black clayey soils, usually above weathering shale.

8. Miscellaneous fragments of shale sandstone and other materials which have absorbed iron and formed a thin polished layer over their surfaces, but which do not strictly enter into this classification.

PLATES

PLATE 1

SOIL PROFILES CONTAINING LAYERS OF CONCRETIONS

FIG. 1. (Left) Profile of a soil of the Eccla series, showing a compact layer of ironstone gravel of type 1a.

FIG. 2. (Right) Profile of a soil of the compact Recent Sands, showing a double concretionary layer of the fourth type.

FIG. 3. Profile of a soil of the compact Recent Sands, showing a thin band of concretions of type 4a.

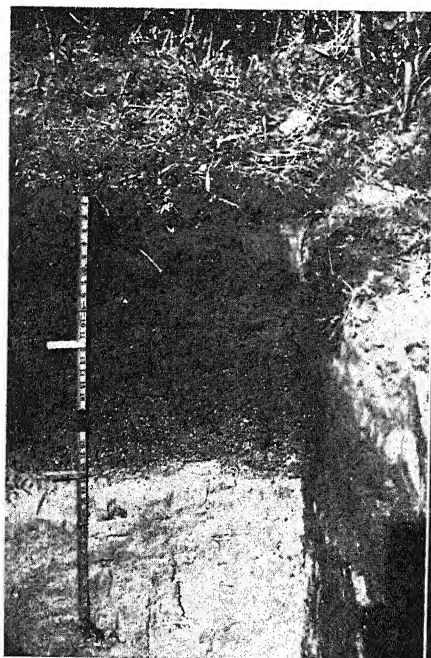


FIG. 1

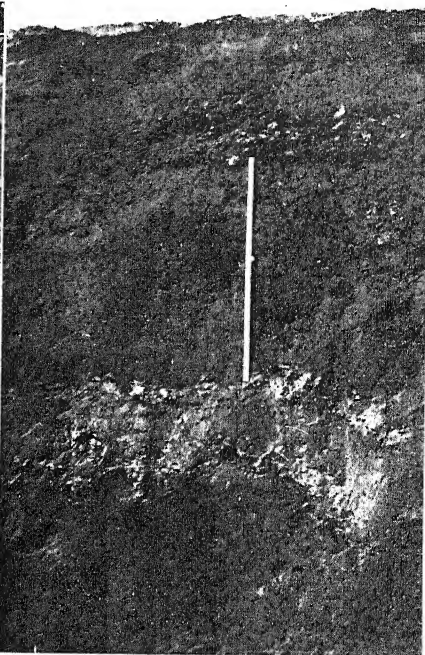


FIG. 2

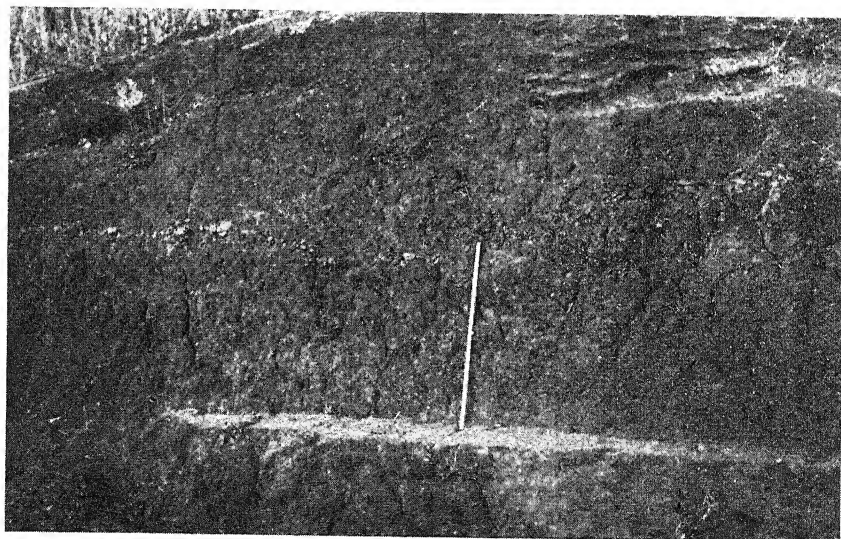


FIG. 3

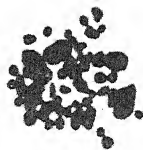
PLATE 2

TYPES OF CONCRETIONS AND REFRACTORY RESIDUES FOUND IN THE NATAL COASTAL SOILS

- 1a. Ironstone gravel (compact layer).
- 1b. Ironstone gravel (loose layer).
2. Rubble (loose layer).
3. Ironstone concretions (throughout profile).
- 4a. Concretions in Recent sands (thin or scattered layer).
- 4b. Concretions in Recent sands (broken cemented layer).
- 5a. Black residue (from seashore).
- 5b. Black residue in Recent sands (throughout profile).
- 5c. Black residue (from Australian coast).
6. Bog iron ore (loose layer).
7. Lime concretions (scattered layer).
8. Miscellaneous concretionary fragments.



1A



1B



2



3



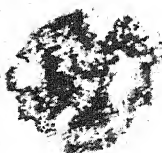
4A



4B



5A



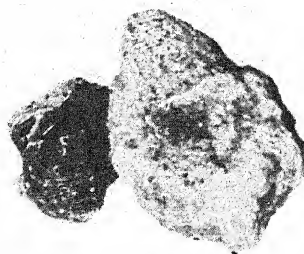
5B



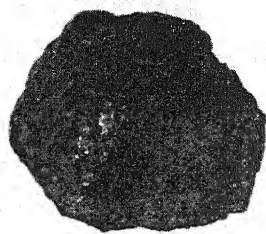
5C



6



7



8

— 0.1 cm.

A MICROSCOPIC METHOD FOR DETERMINING THE WATER-STABLE AGGREGATES IN SOILS¹

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In searching for a means of comparing the natural water-stable structures of soils, a microscopic method has been developed and found useful in characterizing structural differences of wetted soils. The size distribution of aggregates is estimated by direct count, giving an aggregate analysis based on the numbers of particles in each class rather than on their weights. Because of the great number of particles involved, the measurement is based on a sampling method. The aggregates and particles resulting from the slaking of a sample of soil in water are distributed in glycerine, and the resulting suspension is poured into crystallizing dishes where the particles settle according to a random system of distribution. Because of this random distribution small differences in structural stability can be estimated by counting comparatively few microscopic fields.

Severe treatment which would destroy the shapes of the aggregates is not required, since little mechanical action other than that of water slaking is employed and no sieving is necessary. Samples ranging in size from several hundred grams to less than a gram can be used. Natural differences and changes in the morphology of the aggregates can be observed directly.

METHOD

Samples of soil which have been taken with special care to preserve their natural structure are slaked in water in 1-liter Erlenmeyer flasks for 6 hours. The free water is then removed by syphoning, and 97 per cent glycerine is added to make a total volume of 1 liter. The flask is gently rotated until the contents appear to be completely mixed. An aliquot is then quickly poured into a crystallizing dish, and the particles are allowed to settle.

The density of the suspension, which will vary with the different soils used and with the state of division resulting from water slaking, may be controlled by dilution. The best concentration for counting is one that will give a range of particles per field of 0 to 10. For Tama silt loam a concentration of 1 gm. of soil in 500 cc. glycerine has been found suitable for counting aggregates in

¹ A report of part of the work completed on a National Research Fellowship sponsored by the National Research Council. The author is indebted to the University of California for facilities for research, to W. P. Kelley, University of California, for leadership in the work, and to Hans Jenny, University of California, for helpful suggestions.

the range of 1.0 to 0.05 mm. diameter when the suspension is poured into crystallizing dishes to depths of 10 to 20 mm. Within this range of diameters, 100 \times is a suitable magnification. When this soil was shaken in water for an hour, 1 gm. in 1000 cc. was found preferable because of the greater number of small particles formed.

Seventy-millimeter crystallizing dishes are the best size. In smaller dishes the number of fields for counting is limited, and in larger dishes random distribution may be lost through the flowing action of the liquid as it spreads over the dish. Three to five dishes may be poured from each sample, depending on the accuracy required. For most comparisons, counts of 100 fields per crystallizing dish are sufficient. A rectangular field is best, all aggregates touching

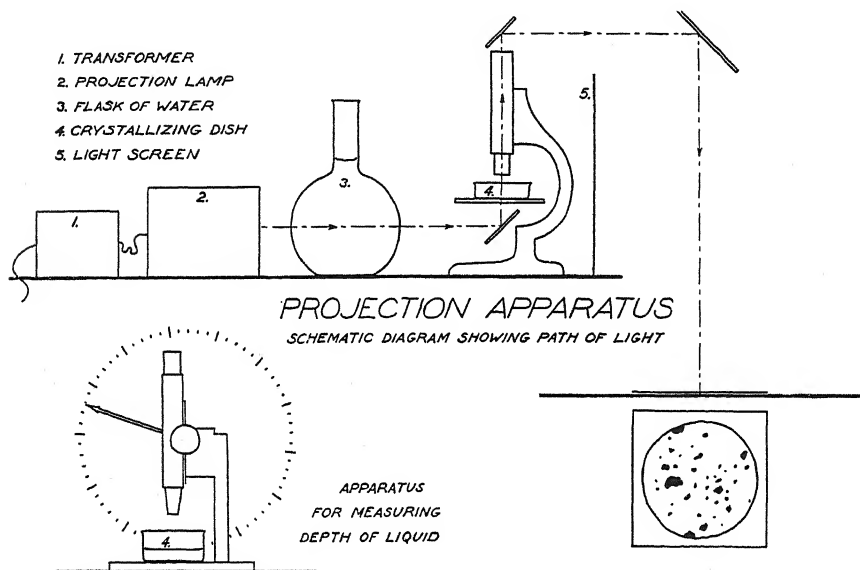


FIG. 1

the right-hand and bottom lines being included and those touching the left-hand and top lines omitted.

A convenient method of measuring the volume of suspension represented by each microscopic field is to determine the area of the field and the depth of the suspension. The latter value can be measured by focusing a microscope on the bottom and on the top of the liquid and reading the vertical displacement on the micrometer uplift screw. Rapid and accurate readings, even on deep suspensions, can be made with a 2- or 3-inch pointer fastened to the large vertical uplift screw and used with a clock-like scale calibrated in microns (fig. 1). Correction must be made for the apparent change in depth of suspension resulting from the differences in indexes of refraction of glycerine and air.

Projection of the shadows of the aggregates on a sheet of coordinate paper will facilitate counting (fig. 1).

The results are expressed as aggregates per gram of air-dry soil.

An analysis of the numbers of ultimate particles in the different size classes selected for aggregate analysis may be desired for comparison with the numbers of water-stable aggregates in each class. Particles as small as 0.002 mm. in diameter can easily be counted. The sample is first prepared as for mechanical analysis determination according to the recommendations of Olmstead et al.² and the number of particles in each class counted as usual.

DISCUSSION

Distribution of aggregates in a dispersing medium

To avoid the necessity of counting so many individuals as to make the procedure impractical, it is necessary to obtain a random arrangement of the aggregates in some medium. Glycerine was found to be a suitable medium. Because of its high viscosity there are no side drift of the aggregates on settling and no stratification of particles before the aliquots can be poured. The aggregates do not tend to group in streaks and swirls in the bottom of the container, as they do when shaken in water. Glycerine is suitable for use with wet soil because it is miscible in water. It is so viscous that little change in the shapes of the aggregates is noticed during the dispersing process. It may be reclaimed by filtering and then heating at about 120°C. until excess water has been driven off.

Glycerine does not cause a measurable effect on aggregation in the size ranges studied, neither does the process of mixing the wet soil with the glycerine cause any noticeable breakdown in structure. This was demonstrated in the measurement of water-stable structure of single natural aggregates selected from the B horizon of Tama silt loam. Two dishes were poured for each aggregate, the samples being shaken between each pouring to remix the suspension. An analysis of the data revealed no significant variability between the results of the first and the second pourings.

To check upon the uniformity of random distribution accomplished by dispersing the aggregates in glycerine and allowing them to settle upon the bottoms of crystallizing dishes, a test was made of the similarity between the observed distributions and those to be expected according to the Poisson curve.³ A good fit was found in almost every case between the actual distribution and the Poisson distribution. This was true for all the size classes studied. It was feared that the larger particles might influence the distribution of smaller ones about them. To test this, counts of smaller aggregates (0.05 to 0.10 mm. in diameter) about larger ones (1 to 2 mm. in diameter) were made, concentric rings of equal area but of increasing distance from the large particle being used as a basis of enumeration. For Tama silt loam at a concentration of 1 gm.

² Olmstead, L. B., Alexander, Lyle T., and Middleton, H. E. 1930 A pipette method of mechanical analysis of soils based on improved dispersion procedure. U. S. Dept. Agr. Tech. Bul. 170.

³ "Student" 1907 On the error of counting with a haemocytometer. *Biometrika* 5: 351-360.

in 500 cc. the distribution of smaller particles in the area covered by these rings was found to follow a random arrangement agreeing closely with the Poisson. Apparently at the concentrations suitable for counting, large particles do not affect the distribution of small ones settling near them.

In most of the studies duplicate samples were used, and three to four crystallizing dishes were poured from each for counting. In many cases the variability between dishes poured from the same sample was so little that two dishes would have been adequate.

The B horizon of Tama silt loam was selected for these studies because of its granular structure and because of the apparent stability of its aggregates in water. It had been noted in the field that erosion deposits of this soil were composed chiefly of aggregates rather than of ultimate particles.

Uniformity of single grains from Tama silt loam, B horizon

By the direct count method it was possible to measure the size distribution of water-stable aggregates in small natural grains of Tama silt loam (table 1).

TABLE 1

Analysis of single aggregates from field samples of the B horizon of Tama silt loam

SAMPLE NUMBER	WEIGHT gm.	AGGREGATES PER GRAM						
		>3 mm.	3-2 mm.	2-1 mm.	1-.5 mm.	.5-.25 mm.	.25-.1 mm.	.1-.05 mm.
1	1.276	0	22	350	1800	8300	40,000	135,000
2	2.357	0	17	300	1500	8100	44,000	154,000
3	1.277	0	17	220	1700	8700	52,000	153,000
4	1.013	0	15	220	1600	8400	49,000	114,000
5	1.207	0	0	200	1600	9100	45,000	118,000

The striking homogeneity of the structural properties of these grains of natural aggregates indicates the intrinsic nature of the forces contributing to the structural characteristics of the horizon from which the samples were taken. Differences between the grains were so slight on the basis of Fisher's⁴ "*F* test" that they could be considered as samples from the same homogeneous population, with the exception of their composition in terms of 0.1 to 0.05 mm. particles. In this size class the difference was not marked, the *F* value being 23.11 as compared with a 5 per cent point of 15.98.

Effect of pretreatment

Comparisons of the natural single-grain aggregates with well-mixed sack samples as ordinarily taken from the field and with samples run through a 2-mm. sieve showed that any process which breaks down the natural dry

⁴ Fisher, R. A. 1935 Statistical Methods for Research Workers. Oliver and Boyd, Edinburgh.

aggregates also breaks down the water-stable aggregates (table 2). Measurements made after slaking intervals of 1, 3, 6, and 12 hours revealed no definite differences in the aggregate analysis of samples from the B horizon of Tama silt loam (table 2).

TABLE 2
Effect of mechanical treatment of sample on size distribution of aggregates*

TREATMENT	AGGREGATES PER GRAM							
	>4 mm.	4-3 mm.	3-2 mm.	2-1 mm.	1-.5 mm.	.5-.25 mm.	.25-.1 mm.	.1-.05 mm.
Single, natural grains, slaked 6 hours	0	0	19	330	1600	8,200	42,700	144,000
Well mixed from sample sack, slaked 1 hour . .	0	0	13	320	2500	13,400	78,000	324,000
Same, slaked 3 hours . .	0	0	30	300	2100	11,700
Same, slaked 6 hours . .	0	0	9	280	1400	12,600
Same, slaked 12 hours . .	0	0	10	280	1700	10,600	77,000	368,000
Sieved through 2-mm. screen, slaked 6 hours.	0	0	1	100	2600	17,000	108,000	215,000

* Sample from B Horizon of Tama silt loam.

TABLE 3
Effect of mechanical treatment of sample on aggregate analysis*

TREATMENT†	AGGREGATES PER GRAM‡							
	>4 mm.	4-3 mm.	3-2 mm.	2-1 mm.	1-.5 mm.	.5-.25 mm.	.25-.1 mm.	.1-.05 mm.
Slaked only	0	0	19	330	1650	8,180	42,000	144,000
Shaken 10 seconds	0	0	7	190	1500	8,160	64,600	253,000
Shaken 30 seconds	2	3	8	150	1860	10,740	99,200	423,000
Shaken 2 minutes	1	1	19	180	1700	8,000	80,300	526,000
Shaken 10 minutes	0	0	4	80	1100	7,000	70,100	534,000
Shaken 30 minutes	0	0	3	40	900	4,520	44,300	710,000
Shaken 1 hour	0	0	0	11	350	2,700	22,700	762,000

* Sample from the B horizon of Tama silt loam.

† All slaked 6 hours in water before shaking. Shaken in 8-oz. bottles, filled to 7 oz., by a reciprocal shaker at rate of 130 complete oscillations per minute.

‡ Averages of two samples.

Ultimate water-stable structure

It is difficult to conceive a method for measuring undisturbed water-stable structure. By extrapolation of curves representing numbers of aggregates per gram plotted against time of shaking in water, an indication, however, may be obtained of how closely the results by this method approach those representing ultimate water-stable structure. Such curves, made from the data in table 3, are shown in figures 2, 3, and 4. These indicate that little effect other than that resulting from the slaking action of the water has taken

place in the natural aggregation of the wet soil studied. From the curves it is also apparent that for the first few minutes of shaking there was a rapid increase in number of aggregates per gram in the size classes smaller than 1 mm., followed by a decline in number in all classes except the 0.1 to 0.05 mm. size as the shaking was continued. The rate of this decline became less as the shaking continued. Apparently there is a state of aggregation which is quite resistant to mechanical action. With prolonged shaking the aggregates

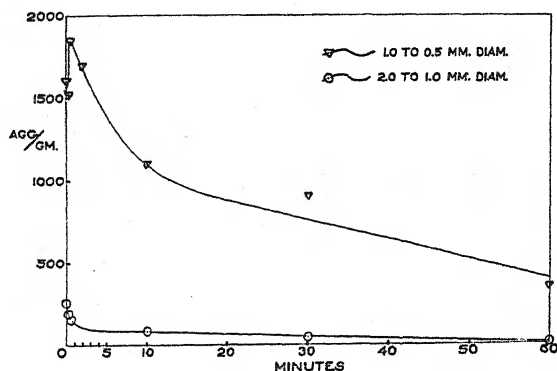


FIG. 2. EFFECT OF TIME OF SHAKING IN WATER UPON NUMBER PER GRAM OF AGGREGATES 1.0 TO 0.5 MM. AND 2.0 TO 1.0 MM. IN DIAMETER

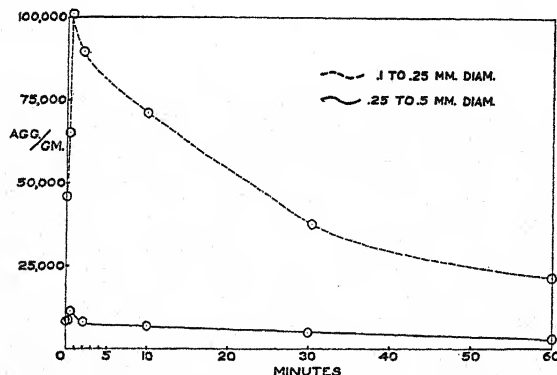


FIG. 3. EFFECT OF TIME OF SHAKING IN WATER UPON NUMBER PER GRAM OF AGGREGATES 0.1 TO 0.25 MM. AND 0.25 TO 0.5 MM. IN DIAMETER

became round and smooth like river gravel, a fact which may have some relation to the marked stability of those aggregates which survive vigorous shaking in water for one-half hour or more.

Measurement of ultimate particles

In order better to evaluate the kind and extent of aggregation it is necessary to know the number of ultimate particles as well as the number of aggregates

which occur in each size class. The size distribution of ultimate particles can be determined by direct microscopic count for all the separates except clay. A count of the number of ultimate particles per gram in the sample of Tama silt loam used for the aggregate analyses reported in this paper revealed no particles larger than 0.1 mm.; 673,000 particles in the class interval the limits of which are 0.1 and 0.05 mm., the limits of very fine sand; and 460,000,000 particles from 0.05 to 0.005 mm. in diameter, the limits of silt. From the data in table 3 it is apparent that in a gram of water-slaked soil there were fewer free particles of the size of very fine sand than there were ultimate particles of that size upon complete dispersion.

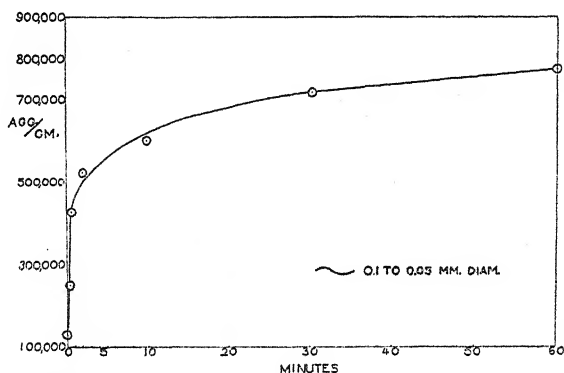


FIG. 4. EFFECT OF TIME OF SHAKING IN WATER UPON NUMBER PER GRAM OF AGGREGATES 0.1 TO 0.05 MM. IN DIAMETER

The following percentages, based on the weights of the different separates found in this soil by mechanical analysis according to Olmstead, et al., are given for comparison with the actual number of particles in each separate:

SEPARATE	PER CENT OF TOTAL WEIGHT	NUMBER PER GRAM
Medium sand.....	0.2	0
Fine sand.....	0.2	0
Very fine sand.....	3.6	673,000
Silt.....	57.7	460,000,000
Clay 0.005 to 0.002 mm.....	5.1	
Clay 0.002 to 0.001 mm.....	4.6	
Clay less than 0.001 mm.....	28.5	
Total.....	99.9	

In order to appraise how closely the numbers found by counting approached a true value, samples of medium sand were counted and the results compared with the expected number per gram of spherical quartz grains with diameters equal to the midpoint of the medium-sand size class, that is, 0.375 mm. The

number of particles by count was 14,800 per gram, whereas the number calculated from the formula $\frac{1}{\frac{1}{6}\pi D^3 P}$, where diameter D is 0.0375 cm. and the density of quartz P is 2.7 gm., is 13,400. Since the sample contained a few mica plates, it would be expected to contain more particles per gram than would a sample of perfect spheres. From this it seems that the values determined by counting are reasonably close to the expected.

If the specific gravity is known, the volume per gram can be calculated. Thus when the number per gram is determined by actual count, the mean volume per particle can be estimated.

SUMMARY

A method for estimating the size distribution of natural water-stable aggregates by direct microscopic count is described. Applicable either to very small or to large samples, the method offers a delicate measure of differences in water-stable structure. That little or no change in the morphology of natural soil aggregates occurs during the analysis is demonstrated.

Analyses of the size distribution of water-stable aggregates in small natural grains of Tama silt loam taken in undisturbed condition from horizon B in the field show a striking similarity among the grains.

A COMPARISON OF THE EFFECT OF CERTAIN CROPPING AND FERTILIZER AND MANURING PRACTICES ON SOIL AGGREGATION OF DUNMORE SILT LOAM¹

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The influence of cropping treatments upon the physical condition of the soil is little understood. Many generalizations on the effect of manure and mineral fertilizers on soil structure have been expressed, but there are few quantitative data in the literature to check the accuracy of these inferences. This paper is an endeavor to evaluate the effect of growing crops, of mineral fertilizers and manure, and of organic matter on the physicochemical properties of the soil which influence aggregation.

This investigation was conducted as a part of the soil and water conservation research program at the Virginia Agricultural Experiment Station, since experimental data (28) had revealed that cropping treatments affect the susceptibility of the soil to erosion. It was felt that soil samples taken from a long-time fertility experiment should show the influence of cropping and fertilizer practices on the physical condition of the soil.

Fertilizer and manure experiments on the College Farm at Blacksburg, Virginia, have been carried on (11) since 1909 on Dunmore silt loam, which has developed from dolomitic limestone. These experiments consist of (a) a 4-year rotation of corn, wheat, clover, and hay, and (b) continuous cropping of corn, wheat, and hay. The hay crop is a grass mixture, predominantly timothy. The continuous cropping plats have been planted to the same crop every year, whereas the rotated plats are arranged in four series, each series being rotated every year. Lime was applied to all plats at 4-year intervals during the experiment, the last application being made in 1936. The pH of the soil does not vary greatly over the experimental area. The treatment of the plats selected for this study is shown in table 1 along with the soil reaction. Each plat represents $\frac{1}{16}$ acre.

EXPERIMENTAL METHODS

After the corn crop was harvested in September, 1939, soil for aggregation studies was taken, by means of a core sampler 3 inches in diameter (20), to

¹ Contribution from the agricultural engineering department, Virginia Agricultural Experiment Station.

² Assistant soil technologist. The author acknowledges assistance and suggestions from J. H. Lillard, Jr., assistant agricultural engineer, and W. Linkous, laboratory assistant.

a depth of 6 inches from three locations on each plat. The soil from the three locations was mixed together, passed through an 8-mm. screen, and stored in a rubber-stoppered jar. Soil aggregation was determined on all samples within 10 days after collection.

The moisture content of the samples was found to range from 13.6 to 22.3 per cent. The soil was kept at field moisture because it appeared that drying

TABLE 1
Treatments and soil reactions of plats

PLAT	TREATMENT PER ACRE (1909-1939)	pH OF PLATS							
		Corn		Wheat		Clover		Hay	
		Check	Ferti- lized	Check	Ferti- lized	Check	Ferti- lized	Check	Ferti- lized
<i>Continuous cropping</i>									
1	16 tons manure once in 4 years; 219 lbs. raw rock phosphate annually	5.3	5.6	5.4	5.6	5.6	6.0
<i>4-Year rotation*</i>									
2	438 lbs. superphosphate annually	5.4	5.1	5.2	5.2	5.0	5.4	5.2	5.2
3	308 lbs. (NH ₄) ₂ SO ₄ , 438 lbs. superphosphate, and 200 lbs. muriate of potash annually	5.2	4.7	5.4	5.2	5.1	4.9	4.9	4.9
6	200 lbs. muriate of potash annually	5.6	6.0	5.6	6.0	5.2	5.1	5.3	5.2
9	308 lbs. (NH ₄) ₂ SO ₄ annually (dried blood used from 1909-1934)	5.0	5.1	5.5	4.5	5.1	5.0	5.0	4.8
11	16 tons manure once in 4 years; 219 lbs. raw rock phosphate annually	5.3	5.7	5.6	5.9	5.3	5.6	5.0	5.5
12	4 tons manure annually	5.5	5.5	5.3	5.6	5.4	5.5	5.6	5.8
13	16 tons manure once in 4 years; 438 lbs. superphosphate annually	5.6	6.2	5.6	6.1	5.6	5.8	5.5	5.5

* Mineral fertilizers—broadcasted on corn land at time of planting, and on wheat, clover, and hay land in spring before growth started.

Manure—When 16 tons were used in a rotation, the manure was applied in December on hay land and plowed under for corn.

When 4 tons were used in a rotation, the manure was applied in December on every plat.

destroyed the aggregating effect of the water films. According to Ehrenberg (7) the simplest form of soil aggregation is the surrounding of soil particles by water films. Henin (10) found that the stability of large aggregates is dependent on the amount of water in the soil. Sideri (22) reported that the surface film creates a definite orientation of the particles in relation to one another. Measurements on the Dunmore soil after a protracted summer drought showed that the field moisture had not fallen below 8 per cent. Air

drying of the same soil in the laboratory, however, reduced the moisture content to 3 per cent.

In evaluating the effectiveness of the wet screening method for measuring soil aggregation, reference is made to the work of Russell and Tamhane (21). These investigators believed that a wet screening method was practically essential if an appreciable proportion of the aggregates was larger than 0.5 mm. They also reported that prescreening the soil allowed for good sampling.

An adaptation of Yoder's (30) wet screening method has been used in this laboratory for measuring soil aggregation. The method employed in this study is described in detail because all aggregate analysis methods are arbitrary. An old "Easy" washing machine was rebuilt for this purpose (plate 1).³ The vertical plunger has been equipped with circular racks for six nests of 5-inch screens. East nest has seven screens with openings 8, 5, 2, 1, 0.5, 0.25, and 0.1 mm. in size. A small lever arm mechanism driven by an electric motor gives a 3.5-inch vertical plunger stroke, and the speed is geared to 36 strokes a minute. Previous to each run the tub was filled with water, and 100 gm. (oven-dry basis) of soil at field moisture was placed on each 5-mm. screen. The 8-mm. screens were placed over them, and the apparatus was operated continuously for 18 minutes. During the 1-6- and the 13-18-minute periods the water was allowed to cover the openings of the 5-mm. screens, and during the 7-12-minute period, to spill over the 8-mm. screens, when the vertical plunger was at its lowest position. The action of the water sorted the aggregates according to size on the various screens. The fraction of very fine sand, silt, and clay not contained in the 5- to 0.1-mm. aggregates, along with the aggregates < 0.1-mm., passed through the bottom screen. At the end of the run the nests were removed from the tub and allowed to drain. The screens were then stacked in an oven to dry, after which they were weighed individually on a small balance. The weight of the dried aggregates represented the percentage of water-stable aggregates in each size group. The percentage of sand > 0.1-mm. was determined by dispersing 100 gm. of the soil with NaOH in an electric mixer and wet sieving through the 0.1-mm. screen. The percentage soil aggregation was calculated as follows:

$$\text{Percentage soil aggregation} = 100 \times \frac{\% \text{ soil aggregates} > 0.1 \text{ mm.} - \% \text{ sand} > 0.1 \text{ mm.}}{100 - \% \text{ sand} > 0.1 \text{ mm.}}$$

Organic matter was calculated from total carbon determinations run by wet oxidation with chromic acid according to Tiurin (27). The soil was passed through a 2-mm. screen before the total soil organic matter was determined. The organic matter in the aggregates retained on the 1- and 0.1-mm. screens was determined and represented "aggregated" organic matter in these sized aggregates.

³ Apparatus designed by J. H. Lillard, Jr.

Soil reaction was determined with a Coleman glass electrode pH meter using a 1:2 soil-water ratio.

Total exchange capacity and exchangeable hydrogen were determined by the barium acetate method as outlined by Prince and Toth (19).

Acid-soluble R_2O_3 , Ca, and Mg were determined in the leachate obtained by treating 25 gm. of soil with 500 cc. of 0.05 *N* HCl (24).

EXPERIMENTAL DATA

The percentage soil aggregation and that of soil aggregates > 1 mm. under the different crops on both the check and the fertilized plots for the con-

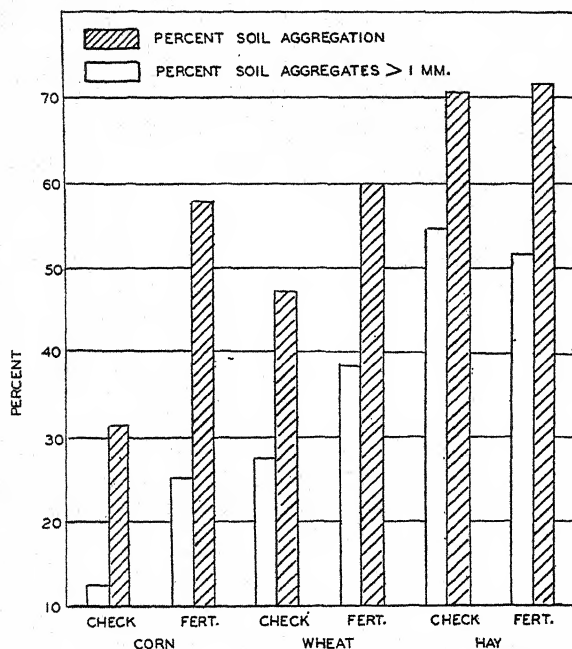


FIG. 1. EFFECT OF FERTILIZER AND CROP ON SOIL AGGREGATION OF CONTINUOUSLY CROPPED PLATS OF DUNMORE SILT LOAM

tinuously cropped plat, no. 1, and the mean of the rotation plats, nos. 2, 3, 6, 9, 11, 12, and 13, are presented graphically in figures 1 and 2. The percentage soil aggregation data for the check and fertilized rotated series are tested for significance in table 2A and 2B. The crops of the check series ranked, Hay = Clover = Wheat = Corn, and those of the fertilized series ranked, Hay = Clover > Wheat = Corn. The data for the soil aggregates > 1 mm. are tested for significance in table 2C and 2D. The check series showed the crops to rank Hay > Clover = Corn > Wheat, and those of the fertilized series ranked Hay = Clover > Corn = Wheat.

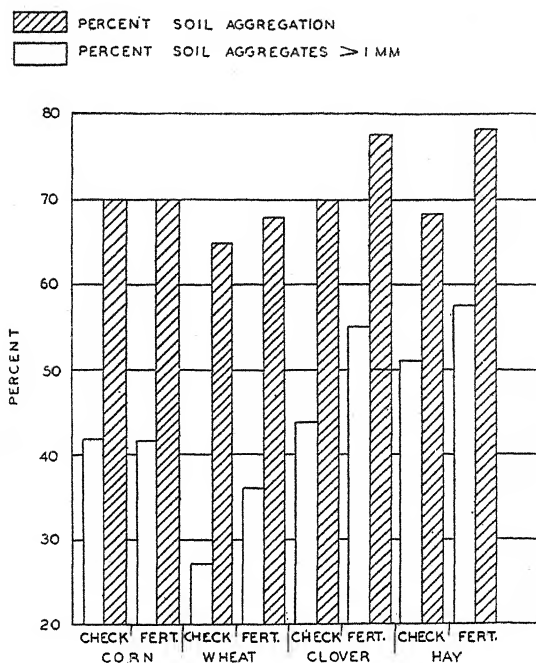


FIG. 2. EFFECT OF FERTILIZER AND CROP ON SOIL AGGREGATION OF CROP ROTATED PLOTS OF DUNMORE SILT LOAM

TABLE 2

Statistical tests by analysis of variance method

	CORN	WHEAT	CLOVER	HAY	SE*
(A) Soil aggregation of rotated check series.....per cent	70.0	65.0	70.3	68.4	6.0
(B) Soil aggregation of rotated fertilized series.....per cent	70.1	68.2	77.7	78.2	6.4
(C) Soil aggregates >1 mm. from rotated check series.....per cent	41.9	29.1	43.9	51.2	6.6
(D) Soil aggregates >1 mm. from rotated fertilized series.....per cent	41.4	33.3	54.7	57.6	8.6
(E) R_2O_3 from soils of rotated check series.....per cent	.25	.29	.25	.17	.067
(F) Ca + Mg from soils of rotated check series.....m.e.	5.98	6.10	5.99	4.49	1.13
(G) Difference between organic matter in total soil and in 1 mm. aggregates—rotated check series.per cent	.35	.32	.30	.20	.086

* Standard error of the difference between means of two crops.

The percentage distribution and the total "relative surface" of the soil aggregates are presented graphically in figures 3 and 4 for the continuous cropping and rotated plots. The total "relative surface" values were calculated from the formula employed by Cole (6):

$$RS = \sum \left(\frac{O_1}{O_1} M_1 + \frac{O_1}{O_2} M_2 + \dots + \frac{O_1}{O_n} M_n \right)$$

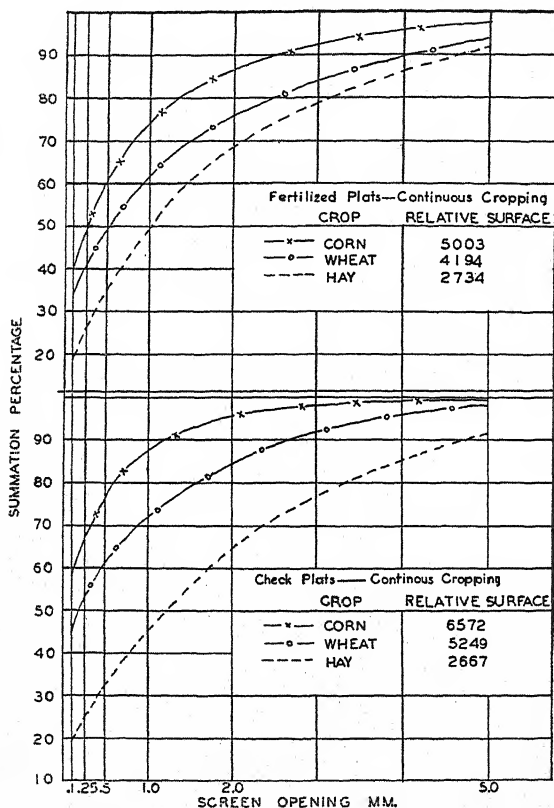


FIG. 3. SIZE DISTRIBUTION OF SOIL AGGREGATES OF CONTINUOUSLY CROPPED PLOTS OF DUNMORE SILT LOAM

where

RS = "relative surface"

O_1 = screen opening of the coarsest screen

O_2 = screen opening of the next coarsest screen

M_1 = percentage of sample retained on the coarsest screen

M_2 = percentage of sample retained on the next coarsest screen

The value used for O_n was 0.05 mm.

Cole (6) pointed out: "The soil aggregates have irregular shapes, yet they are compared as if they had regular shapes. Since their exact dimensions

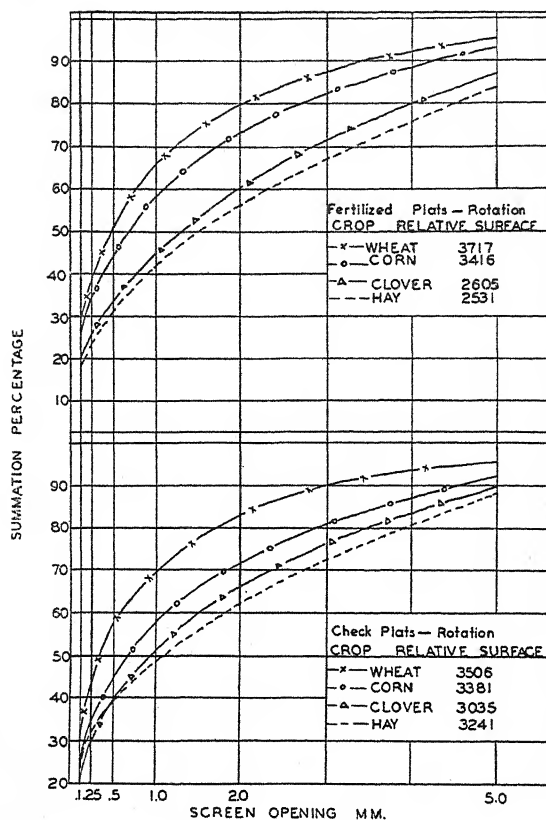


FIG. 4. SIZE DISTRIBUTION OF SOIL AGGREGATES OF CROP ROTATED PLOTS OF DUNMORE SILT LOAM

TABLE 3

R₂O₃, Ca, and Mg leached from soils of check plots with 0.05 N HCl

PLOT	CORN			WHEAT			CLOVER			HAY		
	R ₂ O ₃	Ca	Mg	R ₂ O ₃	Ca	Mg	R ₂ O ₃	Ca	Mg	R ₂ O ₃	Ca	Mg
	per cent	m.e.*	m.e.	per cent	m.e.	m.e.	per cent	m.e.	m.e.	per cent	m.e.	m.e.
1	.21	2.57	1.84	.12	4.51	1.9715	5.81	2.00
2	.24	3.67	1.40	.20	3.33	1.66	.20	5.58	1.49	.09	3.79	1.82
3	.25	3.14	1.60	.26	4.46	1.80	.15	4.79	1.60	.11	2.96	1.26
6	.20	4.57	1.33	.21	3.50	1.40	.27	3.33	1.79	.13	3.50	1.50
9	.27	3.50	1.77	.30	3.89	2.14	.22	3.44	1.40	.20	2.59	1.39
11	.30	5.02	1.88	.38	5.02	1.60	.32	3.50	1.35	.23	2.48	1.04
12	.27	5.40	2.24	.37	4.82	1.73	.29	4.29	1.60	.20	3.44	1.51
13	.21	4.96	1.37	.34	5.75	1.57	.32	5.81	1.94	.24	2.99	1.13
Mean	.25	4.32	1.66	.29	4.40	1.70	.25	4.39	1.60	.17	3.11	1.38

* Milliequivalents per 100 gm. soil.

TABLE 4
R₂O₃, Ca, and Mg leached from soils of fertilized plats with 0.05 N HCl

PLAT	CORN			WHEAT			CLOVER			HAY		
	R ₂ O ₃	Ca	Mg	R ₂ O ₃	Ca	Mg	R ₂ O ₃	Ca	Mg	R ₂ O ₃	Ca	Mg
	<i>per cent</i>	<i>m.e.*</i>	<i>m.e.</i>	<i>per cent</i>	<i>m.e.</i>	<i>m.e.</i>	<i>per cent</i>	<i>m.e.</i>	<i>m.e.</i>	<i>per cent</i>	<i>m.e.</i>	<i>m.e.</i>
1	.35	7.06	2.08	.22	7.38	2.6430	10.89	2.81
2	.35	4.29	1.58	.21	5.87	1.44	.17	8.08	1.91	.23	6.15	1.73
3	.31	3.31	1.93	.22	4.62	1.35	.14	4.23	1.41	.20	3.16	1.23
6	.15	4.51	1.57	.21	3.38	1.46	.23	2.54	1.51	.17	3.16	1.32
9	.27	4.40	2.47	.25	2.09	1.12	.27	2.31	1.32	.29	2.54	1.01
11	.53	10.40	4.09	.44	8.77	2.52	.35	7.50	2.34	.37	5.95	1.75
12	.36	7.00	3.73	.32	6.35	2.79	.23	5.36	2.50	.27	4.68	2.02
13	.42	10.87	4.29	.38	9.19	2.88	.33	6.60	1.78	.35	5.30	1.75

* Milliequivalents per 100 gm. soil.

TABLE 5
Percentage of organic matter in total soil and in soil aggregates

PLAT	CORN			WHEAT			CLOVER			HAY		
	Total	Aggregates		Total	Aggregates		Total	Aggregates		Total	Aggregates	
		1 mm.	0.1 mm.		1 mm.	0.1 mm.		1 mm.	0.1 mm.		1 mm.	0.1 mm.
Check												
1	1.33	1.30	1.14	1.64	1.29	1.29	2.10	1.63	2.67
2	2.43	2.07	2.27	1.93	1.57	1.96	1.76	1.33	2.08	1.55	1.36	2.05
3	2.17	1.73	2.02	2.40	2.20	2.26	1.93	1.50	1.95	1.69	1.45	1.72
6	1.48	1.24	1.17	2.21	1.98	2.21	2.31	2.00	2.21	1.74	1.52	1.19
9	1.64	1.38	1.59	2.00	1.72	1.88	1.88	1.67	1.64	1.55	1.45	1.33
11	2.08	1.71	1.83	2.74	2.36	2.62	2.27	1.98	2.05	1.86	1.64	1.40
12	2.55	2.14	2.50	3.17	2.77	3.03	2.69	2.45	2.62	2.02	1.78	1.86
13	2.21	1.83	1.95	2.76	2.36	2.58	2.93	2.73	2.71	2.21	2.00	1.98
Mean 2, 3, 6, 9, 11, 12, 13	2.08	1.73	1.89	2.46	2.14	2.36	2.25	1.95	2.18	1.80	1.60	1.65
Fertilized												
1	1.93	1.81	2.31	2.34	2.08	2.31	3.00	2.69	3.33
2	2.48	2.33	2.48	2.08	1.81	2.27	1.83	1.24	2.05	2.26	1.76	2.50
3	2.55	2.74	2.76	2.36	2.14	2.48	2.07	1.71	2.31	2.43	1.98	2.71
6	1.83	1.86	1.36	2.19	2.24	2.02	2.50	2.14	2.52	1.95	2.12	1.93
9	2.14	1.88	2.05	1.98	1.55	1.79	2.31	2.07	2.00	2.08	2.02	2.00
11	3.74	3.00	3.33	3.24	2.76	2.77	2.88	2.88	2.79	2.83	2.65	2.38
12	3.62	3.07	3.43	3.27	3.14	3.24	3.07	2.83	3.20	2.95	2.55	2.62
13	4.07	3.27	3.58	3.50	3.26	3.22	3.07	3.02	2.98	3.00	3.07	3.02
Mean 2, 3, 6, 9	2.25	2.20	2.16	2.15	1.94	2.14	2.18	1.79	2.22	2.18	1.97	2.29
Mean 11, 12, 13	3.81	3.11	3.45	3.34	3.05	3.08	3.01	2.91	2.99	2.93	2.76	2.67

are not known, the calculation of the surface cannot be made in definite units, but only in relative terms, and so the comparison of surfaces on all the fractions of the sample is called 'relative surface' and is expressed in non-dimensional units."

The percentage R_2O_3 and the milliequivalents of Ca and Mg removed from the soil by leaching with 0.05 *N* HCl are shown in tables 3 and 4 for the check and the fertilized series. The data for the rotated check series are tested for significance in table 2E and 2F. For both percentage R_2O_3 and milliequivalents Ca + Mg, the crops ranked Hay < Clover = Corn = Wheat.

The percentages of organic matter in the total soil, in the 1-mm. aggregates, and in the 0.1-mm. aggregates are shown in table 5. Relative differences were determined by subtracting the percentage of organic matter in the 1-mm. aggregates from that in the total soil. The differences for the rotated check series are tested for significance in table 2G. The crops ranked Hay > Clover = Corn = Wheat for the amount of the total soil organic matter which was present in the 1-mm. aggregates.

TABLE 6
Cation exchange capacity and exchangeable hydrogen in soil

PLAT	TREATMENT	CORN			HAY		
		Exchange Capacity	Exchangeable H		Exchange Capacity	Exchangeable H	
		<i>m.e.*</i>	<i>m.e.</i>	<i>per cent</i>	<i>m.e.</i>	<i>m.e.</i>	<i>per cent</i>
11	Fert.	12.42	5.00	40.3	7.67	3.79	49.4
12	Fert.	12.50	5.00	40.0	8.68	3.69	42.5
13	Fert.	14.18	3.75	26.4	9.15	4.60	50.3
Mean.....		13.03	4.58	35.6	8.50	4.03	47.4

* Milliequivalents per 100 gm. soil.

The total base exchange capacity, exchangeable hydrogen, and per cent hydrogen saturation for some of the plat soils are shown in table 6.

DISCUSSION

Methods of expressing soil aggregation

The soil aggregates are the natural fragments into which a soil breaks up. These aggregates are collections of primary soil particles and behave as independent particles. In this paper two methods of expressing the aggregate condition of the soil have been employed. These are (a) the percentage soil aggregation, and (b) the size distribution of the soil aggregates and "relative surface."

The percentage soil aggregation, represented in figures 1 and 2, shows the effect of the crop, fertilizer and manure, and cropping system on the aggregate condition of the soil. The continuously cropped plats showed the soil of the

fertilized wheat plats and corn plats to have a higher percentage soil aggregation than the corresponding check plats. The percentage soil aggregation was the same for the check and fertilized rotated corn and wheat plats. The rotated fertilized clover and hay plats showed an increase in the percentage soil aggregation over the corresponding check plats. The percentage soil aggregation ranked the crops of the rotated check series as follows, Hay = Clover = Corn = Wheat. The data for the aggregates > 1 -mm., however, ranked the crops Hay $>$ Clover = Corn $>$ Wheat. This is an important consideration because Krause (12) has pointed out that the work of the Russians indicated that an aggregate diameter of 1-3 mm. was the optimum aggregate size for the best distribution of air and water in the soil.

The term "relative surface" as applied to tilth has a physical meaning. The relative surface values are used to compare the effect of the crop, fertilizer and manure, and cropping system on soil aggregation. The "relative surface" values depend on the size distribution curves, and the curves occurring higher on the graphs in figures 3 and 4 represent a soil condition with small aggregates having a high total relative surface. Under continuous cropping, the soil of the corn plats, both fertilized and check, had the highest total "relative surface," and that of the hay plats, the lowest. In the rotation experiment, the total "relative surface" of the soil was highest in the wheat plats, both fertilized and check, lower in the corn plats, and lowest in the clover and hay plats. Comparison of the check and fertilized rotation plats for the individual crops showed that the soil of the corn plats had the same total "relative surface," the soil of the fertilized wheat plats had a higher "relative surface" than the check, and the soil of the fertilized clover and hay plats had a much lower total "relative surface."

Organic matter and soil aggregation

Several investigators (1, 2, 25, 26) have shown that a relationship does exist between soil organic matter and soil aggregation, whereas others (3, 5) have reported that additions of organic matter may or may not affect the aggregation of the soil. The present study showed that on the continuously cropped corn and wheat plats, an increase in the organic matter content of the fertilized and manured plats was accompanied by an increase in the percentages of soil aggregation and of aggregates > 1 -mm. over those of the corresponding checks (fig. 1). The fertilized and rotated corn and wheat plats contained more organic matter than the check plats but did not show any increase in percentage soil aggregation over the corresponding checks (fig. 2).

Metzger and Hide (16) reported that the more aggregated portions of the soil contained more organic matter. In this study the 1-mm. aggregates of the rotated check hay plats contained more of the total organic matter than did the 1-mm. aggregates of the clover, corn, or wheat plats. The check hay plats also had a higher percentage of aggregates > 1 -mm. than did any of the other three crops in this series. The 0.1-mm. aggregates of the corn, wheat,

and clover check rotated plats contained more organic matter than did the corresponding 1-mm. aggregates. The difference was insignificant for the hay plats.

Geltzer (9) suggested that the amelioration in structure under an established grass or clover sod may be caused by the rapid decomposition of the large supply of readily decomposable organic substances. The explanation may be that during the process of mineralization of the organic matter in the soil there was an accumulation of synthetic microbial substances which brought about the formation of large soil aggregates. A greater rate of decomposition of the organic matter should result in a larger amount of these substances with better soil aggregation. In investigating the effect of soil microorganisms on aggregation, Martin and Waksman (13) reported that when complex organic materials were used as a source of energy for the microorganisms, the more readily the material was destroyed, the greater was the binding action on the soil particles. The present study showed that the check rotated hay plats contained less total organic matter than did any of the other check plats. The soil of the hay plats also had a higher percentage of aggregates > 1-mm. than did any of the other three crops in this series.

On the fertilized rotation series manure was applied immediately before corn on plats 11, 12, and 13. The total soil contained 22 per cent more organic matter than did the 1-mm. aggregates from the corn plats. This difference was greatly reduced on the manured clover and hay plats, which had about the same amount of organic matter in the total soil and in the 1-mm. aggregates. The hay crop left more organic matter tied up in the 1-mm. aggregates than did corn, wheat, or clover when the soil was untreated. On the fertilized series the same effect was manifested under the clover crop, or 1 year earlier than in the check rotation. It may be reiterated that the aggregates > 1-mm. ranked the crops Hay > Clover = Corn > Wheat for the check rotation series, and Hay = Clover > Corn = Wheat for the fertilized rotation.

Physicochemical relationships as affecting soil aggregation

Myers (17) has explained the specific effect of humus in soil aggregation as the formation of a chemical union between the organic and inorganic colloids, especially under acid conditions; the mixing of these colloids resulted in a reduction of the cation exchange capacity of the systems from the calculated values. Mattson (14, 15) suggested that loss in exchange capacity of humus acidoid may be due to a combination with Al or Fe. Boratynski and Mattson (4) asserted that such a combination may also increase the strength of the humus acidoid by causing H ions to dissociate below pH 7 which in the free acidoid do not dissociate. The present study showed that the soil from the manured rotated hay plats had a lower cation exchange capacity and a higher percentage of hydrogen than the soil of the corn plats (table 6). The difference in cation exchange capacity between these plats may be attributed to the higher percentage of organic matter in the soil of the corn plats. While the

soil of the corn plats had 27 per cent more organic matter than that of the hay plats, it had 53 per cent greater exchange capacity. This was associated with less soluble R_2O_3 from the soil of the hay plats.

Much of the experimental evidence on the effect of lime on soil structure is still contradictory. The general opinion is that the presence of lime in the soil or the degree of calcium saturation affects soil aggregation only by hastening the decomposition of the organic matter. Martin and Waksman (13) reported experimental data to confirm this opinion. Peele (18) and Elson and Lutz (8) reported that liming Cecil sandy loam resulted in a decrease in soil aggregation. In this investigation the soil of the check hay plats, with less $Ca + Mg$ than that of the clover, wheat, or corn plats, had a higher percentage of aggregates > 1 -mm.

In studying clay-humus formations, Sideri (23) reported that the elimination of iron and aluminum oxides from the surface of clay particles increased the ability of the particles to aggregate. He believed that the presence of large amounts of these oxides destroyed the orienting properties of clay with respect to humus. Rogers (20) reported that increasing quantities of soluble aluminum in the soil were accompanied by decreasing quantities of silt and clay aggregated into units larger than silt size. In the present study the percentage R_2O_3 and the milliequivalents $Ca + Mg$ removed from the soil of the rotated check plats by 0.05 N HCl ranked the crops Hay $<$ Clover = Corn = Wheat. Waksman (29) pointed out that in the absence of the divalent cations, humus combined with R_2O_3 or clay fractions. The amount of the total soil organic matter present in the 1-mm. aggregates ranked the crops Hay $>$ Clover = Corn = Wheat, and for the soil aggregates > 1 -mm. the crops ranked Hay $>$ Clover = Corn $>$ Wheat. The fact that there was less $Ca + Mg$ in the soil under the hay crop can be explained by the removal of these cations by the crop. The decreased solubility of the R_2O_3 in the soil under the hay crop may be explained by a firmer bond with the organic colloids. This same general tendency was evident on the fertilized rotation series, but no statistical analysis was attempted because of the effect of the fertilizer and manure in increasing the organic matter content and the Ca and Mg in the soil.

Effect of the growing crop on soil aggregation

An important consideration in the formation of water-stable aggregates in the soil was the length of time the soil remained undisturbed. Russian workers, who followed the effect of pasture on the structure of the soil, found that 2 or 3 years was necessary to produce any noticeable improvement. In this study the soil of the check rotated hay plats had not been disturbed since the wheat crop was seeded, 2 years earlier in the rotation. The higher percentage of aggregates > 1 -mm. on these plats is an indication that the formation of soil aggregates is a dynamic process. Cultivation of the soil for the corn and wheat crops destroyed the aggregates > 1 -mm., but the 4-year rotation allowed the large aggregates to form again when the soil was left un-

disturbed in clover and hay. The fertilizing and manuring of the crops in the 4-year rotation so hastened the formation of the large aggregates that the soil of the clover plats had the same percentage of these aggregates as the corresponding hay plats (fig. 2). The improvement in soil structure was caused by a combination of physicochemical changes, one of which was the greater rate of decomposition of organic matter in the soil under the clover and hay crops.

SUMMARY

The effect of crops, fertilizer and manure, and cropping systems on soil aggregation of Dunmore silt loam from long-time fertility experiments at Blacksburg, Virginia, was investigated in the laboratory. A specially designed wet screening apparatus was used for measuring soil aggregation.

It was found that the size distribution curve of the soil aggregates and "relative surface" gave a more complete picture of the aggregate condition of the soil than did the percentage soil aggregation.

The continuous growing of corn or wheat on the soil reduced both the percentage soil aggregation and that of aggregates > 1 -mm. The manuring of continuously cropped corn or wheat plats resulted in an increase in soil aggregation over the corresponding check plats.

The soil of plats that had been under hay for 30 years was better aggregated than that of plats continuously cropped to corn or wheat.

It was found that the percentage soil aggregation remained the same during a 4-year rotation of corn, wheat, clover, and hay when no manure or fertilizer was applied to the soil. The soil of the hay plats contained a higher percentage of aggregates > 1 -mm. This indicated that soil aggregation was a dynamic process, with large aggregates forming in the soil when it was allowed to remain in sod for several years. In the same 4-year rotation, when all crops were fertilized or manured, the soil of the clover and hay plats had a higher percentage of soil aggregation and of aggregates > 1 -mm. than the soil under corn or wheat.

The soil of plats that had been in sod for 30 years was as well aggregated as the soil of fertilized or manured plats of clover or hay in a 4-year rotation.

The physicochemical data showed that the soil under the rotated manured corn plats had a greater exchange capacity and a lower percentage hydrogen saturation than did the soil under the hay plats in the same rotation.

The soil of rotated check hay plats had a higher percentage of the total soil organic matter tied up in the 1-mm. aggregates than did the soil from rotated check corn, wheat, or clover plats. The soil of the hay plats also had less organic matter than that of any of the other plats.

Organic matter applied in the form of stable manure directly to corn land was not tied up completely with the 1-mm. aggregates.

The soil of the rotated check hay plats contained less Ca and Mg and soluble R_2O_3 than the soil from corn, wheat, or clover plats.

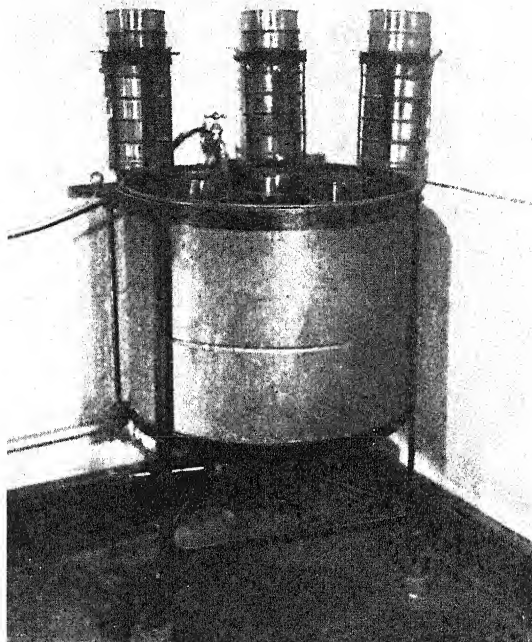
REFERENCES

- (1) BAVER, L. D. 1935 Factors contributing to the genesis of soil microstructure. *Proc. Amer. Soil. Survey Assoc. Bul.* 16: 55-56.
- (2) BAVER, L. D., AND RHOADES, H. F. 1932 Aggregate analysis as an aid in the study of soil structure relationships. *Jour. Amer. Soc. Agron.* 24: 920-930.
- (3) BERTRAMSON, B. R., AND RHOADES, H. F. 1938 The effects of cropping and manure applications on some physical properties of a heavy soil in eastern Nebraska. *Proc. Soil Sci. Soc. Amer.* 3: 32-36.
- (4) BORATYNSKI, K., AND MATTSON, S. 1938 The electrochemistry of soil formation: III. The humate complex. *Ann. Agr. Col. Sweden* 7: 63-119.
- (5) BROWNING, G. W. 1937 Changes in the erodibility of soils brought about by the application of organic matter. *Proc. Soil Sci. Soc. Amer.* 2: 85-86.
- (6) COLE, R. C. 1939 Soil macrostructure as affected by cultural treatments. *Hilgardia* 12: 429-472.
- (7) EHRENBERG, P. 1931 Neue Fragen der Bodenstruktur. *Ztschr. Pflanzenernähr., Düngung, und Bodenb.* (A) 19: 44-77.
- (8) ELSON, J., AND LUTZ, J. F. 1940 Factors affecting aggregation of Cecil soils and the effect of aggregation on run-off and erosion. *Soil Sci.* 50: 265-275.
- (9) GELTZER, F. U. 1937 The role of organic matter in the formation of soil structure. *Chemization Socialistic Agr. (U.S.S.R.)* 7: 45-61; 8: 53-63.
- (10) HENIN, S. 1938 Étude Physico-Chimique de la Stabilité Structurale des Terres. Imprimerie Nationale, Paris.
- (11) KIPPS, M. S., AND HUTCHESON, T. B. 1938 Fertilizer and manure experiments on Dunmore Silt loam soil. Va. Polytech. Inst. Bul. 317.
- (12) KRAUSE, M. 1931 Russische Forschungen auf dem Gebiete der Bodenstruktur. *Landw. Jahrb.* 73: 603-690.
- (13) MARTIN, J. P., AND WAKSMAN, S. A. 1940 Influence of microorganisms on soil aggregation and erosion. *Soil Sci.* 50: 29-47.
- (14) MATTSON, S. 1931 The laws of soil colloidal behavior: IV. Isoelectric precipitates. *Soil Sci.* 31: 55-77.
- (15) MATTSON, S. 1931 The laws of soil colloidal behavior: V. Ion absorption and exchange. *Soil Sci.* 31: 311-331.
- (16) METZGER, W. H., AND HIDE, J. C. 1938 Effect of certain crops and soil treatments on soil aggregation and the distribution of organic matter in relation to aggregate size. *Jour. Amer. Soc. Agron.* 30: 833-843.
- (17) MYERS, H. E. 1937 Physicochemical reactions between organic and inorganic soil colloids as related to aggregate formation. *Soil Sci.* 44: 331-359.
- (18) PEELE, T. C. 1937 The effect of lime and organic matter on the erodibility of Cecil clay. *Proc. Soil Sci. Soc. Amer.* 2: 79-84.
- (19) PRINCE, A. L., AND TOTH, S. J. 1936 The effect of phosphates on the cation exchange capacity of certain soils. *Soil Sci.* 42: 281-290.
- (20) ROGERS, H. T. 1939 The relation between soil reaction, erosion, and aggregation of silt and clay in Clarksville loam. *Jour. Amer. Soc. Agron.* 31: 915-925.
- (21) RUSSELL, E. W., AND TAMMANE, R. V. 1940 The determination of the size distribution of soil clods and crumbs. *Jour. Agr. Sci.* 30: 210-234.
- (22) SIDERI, D. I. 1936 On the formation of structure in soil: II. Synthesis of aggregates; on the bonds uniting clay with sand and clay with humus. *Soil Sci.* 42: 461-479.
- (23) SIDERI, D. I. 1938 On the formation of structure in soil: IV. The structure of mixed clay-sand and clay-humus formations. *Soil Sci.* 46: 129-137.
- (24) SLATER, C. S., AND BYERS, H. G. 1934 Base exchange and related properties of the colloids of soils from the Erosion Experiment Stations. U. S. Dept. Agr. Tech. Bul. 461.

- (25) SOKOLOWSKY, A. N. 1933 The problem of soil structure. *Trans. First. Conn. Internatl. Soc. Soil Sci. (Soviet Sect. Moscow)* (A) 1: 34-110.
- (26) TIULIN, A. 1933 Certain considerations on the genesis of soil structure and on methods for its determination. *Trans. First. Conn. Internatl. Soc. Soil Sci. (Soviet Sect. Moscow)* (A) 1: 111-132.
- (27) TIURIN, I. V. 1931 New modification of the volumetric method of determining soil organic matter by means of chromic acid. *Pedology* 28: 36-47.
- (28) Virginia Agricultural Experiment Station 1940 Soil and water conservation investigations 1936-1939. *Prog. Rpt. Agr. Engin. Dept.*
- (29) WAKSMAN, S. A. 1938 *Humus*, ed. 2. Williams & Wilkins Co., Baltimore.
- (30) YODER, R. E. 1936 A direct method of aggregate analysis of soils and a study of the physical nature of erosion losses. *Jour. Amer. Soc. Agron.* 28: 337-351.

PLATE 1

WET SCREEN APPARATUS FOR DETERMINING WATER-STABLE SOIL AGGREGATES



NITRATE FERTILIZER ADDITIONS TO WATERLOGGED SOILS IN RELATION TO OXYGEN DEFICIENCY¹

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The importance of an adequate oxygen supply for the vital activities of plant roots is well known (2, 4). In the growth of fruit trees, however, the maintenance of favorable aeration conditions in certain soils is difficult. There are many areas in the fruit-growing regions of California in which a high water table persists during certain seasons of the year, and on some soils the management of irrigation practice so as to avoid temporary waterlogging is not easy. In the tropics this problem is also acute. The control of moisture in some of these regions is almost impossible because of very heavy rainfall during certain periods of the year. In areas of heavy soils, saturation and consequently lack of aeration may exist for months at a time. Where impervious subsoils are encountered, a temporary water table which persists for some months may be formed within the area of the root zone.

The findings of Arnon (1) and Haas (3), that lack of oxygen in solution cultures apparently can be offset in some measure by an adequate nitrate supply, have raised the question whether the presence or application of nitrate fertilizer would in any way minimize the ill effects of oxygen deficiency in soil. In order to test this question, soil experiments in pots employing grapefruit and avocado seedlings were carried out.

EXPERIMENTAL

The soil used in these experiments was an unfertilized Ramona loam. Though no nitrate or ammonia determinations were made at the outset, previous studies with this soil have shown it to be decidedly low in both total and available nitrogen.

Individual avocado and grapefruit seedlings were grown in 3-gallon pots of soil for an initial period of about 3 months under uniform conditions in the greenhouse to get them well established. At the end of this time a series of differential fertilizer and watering treatments were begun. The grapefruit

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seedlings were transplanted into the pots on September 18, 1938, and the differential treatments were begun on December 30. Avocado seeds of the Mexicola variety were planted on September 14-16, 1938, and the treatments were begun on January 9, 1939. The grapefruit seedlings were fairly uniform; except for one or two pots their heights at the start of differential treatments averaged about 20 inches. The avocados were less uniform in growth, with heights ranging from 6 to 22 inches. The replicates of the treatments with avocados were grouped so that in any one series of comparisons plants of about the same growth and vigor were used.

The grapefruit and avocados were divided into four groups of six pots each, and the following watering treatments were instituted: A, normal watering, free drainage;⁴ B, excess watering, i.e., soil kept continuously wet, free drainage; C, waterlogged, i.e., an inch or more of free water maintained continuously on the soil surface by plugging the drainage hole in the bottom of the pot; and D, normal watering, no drainage. Distilled water was used throughout. In each group two pots were given $(\text{NH}_4)_2\text{SO}_4$; two, $\text{Ca}(\text{NO}_3)_2$; and two were left untreated. The chemicals were applied in solution at a rate equivalent to 150 pounds nitrogen per acre. The pot numbers and treatments accorded the grapefruit series are shown in table 1. A similarly numbered series with identical fertilizer and water treatments was set up for the avocados.

The experiment was carried on in this manner for about $2\frac{1}{2}$ months. No cultures in which there was free drainage showed ill effects, even though one group was excessively watered. Likewise, no injury occurred in the group without free drainage, on which water was applied in amounts just sufficient to meet plant requirements.

In the waterlogged soils, however, marked effects occurred. Though the behavior of the individual plants in duplicate pots was not entirely uniform, wilting of the avocado and leaf yellowing (plate 2) of the citrus plants, symptoms which commonly develop on field trees injured by excessive water, became manifest in from 10 to 20 days after waterlogging was commenced. The avocado plants treated with $\text{Ca}(\text{NO}_3)_2$ were the first to be affected, followed in order by the $(\text{NH}_4)_2\text{SO}_4$ cultures and the nonfertilized plants. The grapefruit seedlings growing in the soils treated with $(\text{NH}_4)_2\text{SO}_4$ were the first to be affected, the $\text{Ca}(\text{NO}_3)_2$ and the control cultures behaving more or less similarly. The size of avocado and grapefruit plants used in the experiment and some of the adverse effects of the nitrogen-fertilizer treatments under conditions of waterlogging are shown in plate 1.

Nitrate determinations were made on drainage water drawn from the waterlogged pots of both series on January 23 and again on February 27. The results (table 2) show that when the first effects of overwatering became manifest some nitrate was present. The second series of drainage samples taken on February 27 were nearly all devoid of nitrate.

⁴By "normal watering" is meant supplying water as needed. In practice, the pots were thoroughly watered, then allowed to dry to somewhere near the wilting point, and the process was repeated.

This experiment was discontinued on March 17, 1939. The injured plants were harvested and the roots examined. Severe root rotting had occurred in all the water-saturated cultures.

TABLE 1
Treatments accorded grapefruit seedlings in 3-gallon pots at beginning of experiment

GROUP	POT NUMBER	FERTILIZER TREATMENT	WATERING
A	1	Control	Normal watering, free drainage
	2		
	3	(NH ₄) ₂ SO ₄	
	4		
	5	Ca(NO ₃) ₂	
	6		
B	7	Control	Excess watering, free drainage
	8		
	9	(NH ₄) ₂ SO ₄	
	10		
	11	Ca(NO ₃) ₂	
	12		
C	13	Control	Waterlogged (The drainage hole in the bottom of the pot was plugged with a cork stopper and an inch or more of free water maintained on the soil surface)
	14		
	15	(NH ₄) ₂ SO ₄	
	16		
	17	Ca(NO ₃) ₂	
	18		
D	19	Control	Normal watering; drainage hole in bottom of pot stoppered
	20		
	21	(NH ₄) ₂ SO ₄	
	22		
	23	Ca(NO ₃) ₂	
	24		

Inasmuch as the nonwaterlogged grapefruit and avocado plants (groups A, B, and D) were still entirely healthy, it was decided to repeat the fertilizer treatment on all groups and saturate groups A and D with water, utilizing the B group for controls. Those pots of all series which had received previous treatments of $(\text{NH}_4)_2\text{SO}_4$ and $\text{Ca}(\text{NO}_3)_2$, respectively, were retreated with the

same fertilizers, at the following rates: Group A received amounts equivalent to 100 pounds N per acre; group B, 200 pounds; and group D, 400 pounds. The nitrogen fertilizers were applied in solution as before; one-half of the nitrogen was added on April 15, and the remainder on April 17, 1939. Enough solution was applied on this latter date to promote a small amount of leaching in all pots. These leachings were caught and analyzed for nitrate and nitrite. The results of the analyses are shown in table 3. The drainage water from pots receiving no fertilizer contained only traces of nitrate and nitrite. Those pots which had received $(\text{NH}_4)_2\text{SO}_4$ or $\text{Ca}(\text{NO}_3)_2$ at the outset and again on

TABLE 2
Nitrate nitrogen in drainage waters from waterlogged soils

DATE	PLANT	POT NUMBER	TREATMENT*	$\text{NO}_3\text{--N}$
1/23/39 (24 days after waterlogging commenced)	Grapefruit			<i>p.p.m.</i>
		13	None	0.0
		14	None	0.25
		15	$(\text{NH}_4)_2\text{SO}_4$	0.0
		16	$(\text{NH}_4)_2\text{SO}_4$	4.00
		17	$\text{Ca}(\text{NO}_3)_2$	2.40
		18	$\text{Ca}(\text{NO}_3)_2$	4.00
2/27/39	Grapefruit	14	None	0.0
		16	$(\text{NH}_4)_2\text{SO}_4$	0.0
		18	$\text{Ca}(\text{NO}_3)_2$	0.0
1/23/39 (14 days after waterlogging commenced)	Avocado	13	None	22.40
		14	None	0.25
		15	$(\text{NH}_4)_2\text{SO}_4$	16.00
		16	$(\text{NH}_4)_2\text{SO}_4$	0.60
		17	$\text{Ca}(\text{NO}_3)_2$	160.00
		18	$\text{Ca}(\text{NO}_3)_2$	200.00
2/27/39	Avocado	14	None	0.0
		16	$(\text{NH}_4)_2\text{SO}_4$	0.0
		18	$\text{Ca}(\text{NO}_3)_2$	8.00

* The grapefruit cultures were treated with the nitrogen fertilizers on December 30, 1939, and the avocados on January 9. Waterlogging was commenced on these dates.

April 15 and April 17 showed varying amounts of nitrate and nitrite. It is clear that the soil solution of the pots receiving nitrate fertilizer contained a substantial supply of nitrate at the time waterlogging was begun.

Following the sampling of drainage on April 17, the drainage holes of pots in groups A and D were stoppered, and enough additional water was applied to flood the pots to a depth of $1\frac{1}{2}$ inches. Thenceforth until the termination of this second experiment, the soil surface was kept covered with water. A piece of roofing paper fitted around the plant was placed on the surface of each pot to prevent algal growth.

On April 21, four days after the saturation of pots in groups A and D, the temperature in the greenhouse reached a peak of 100°F. Though none of the citrus plants were affected, all but one of the waterlogged avocado plants showed wilt. The plants in the pots receiving nitrogen were more adversely affected than those which received no nitrogen. The avocados in pots 23 and 24 were severely affected. It is possibly significant that, in these, nitrite nitrogen was high (30 p.p.m.). In sharp contrast to the avocados growing in the water-saturated soils, those not waterlogged, irrespective of treatment, showed no wilt.

TABLE 3
Nitrate and nitrite nitrogen in drainage from treated and untreated pots

GROUP	POT NUMBER	FERTILIZER TREATMENT*		GRAPEFRUIT		AVOCADOS	
		Kind	Rate per acre	NO ₃ -N	NO ₂ -N	NO ₃ -N	NO ₂ -N
			<i>lbs.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>	<i>p.p.m.</i>
A	1	None	...	Trace	Trace	Trace	Trace
	2	None	...	Trace	Trace	Trace	Trace
	3	(NH ₄) ₂ SO ₄	100	85	Trace	10.00	12.00
	4	(NH ₄) ₂ SO ₄	100	135	15.00	160.00	2.00
	5	Ca(NO ₃) ₂	100	70	8.00	50.00	12.00
	6	Ca(NO ₃) ₂	100	115	3.80	210.00	1.60
B	7	None	...	None	Trace	Trace	0.06
	8	None	...	None	Trace	Trace	0.10
	9	(NH ₄) ₂ SO ₄	200	None	Trace	27.0	0.24
	10	(NH ₄) ₂ SO ₄	200	None	Trace	Trace	Trace
	11	Ca(NO ₃) ₂	200	140	12.80	42	Trace
	12	Ca(NO ₃) ₂	200	120	3.20	175	Trace
D	19	None	...	None	Trace	Trace	0.14
	20	None	...	0.5	Trace	Trace	0.14
	21	(NH ₄) ₂ SO ₄	400	1.7	0.05	†	†
	22	(NH ₄) ₂ SO ₄	400	1.3	1.00	4.00	2.00
	23	Ca(NO ₃) ₂	400	325.0	36.00	85.00	30.00
	24	Ca(NO ₃) ₂	400	375.0	30.00	350.00	30.00

* The nitrogen fertilizers were applied on April 15 and 17, 1939, and the leachings collected and analyzed on the latter date.

† The plant in the pot was too poor to be included in the experiment.

The fact that the grapefruit seedlings were uninjured indicates that they may be less sensitive to oxygen deficiency or the secondary effects resulting therefrom than are avocados.

Two days later, on April 23, when the greenhouse temperatures became more nearly normal, those avocados which were only slightly affected began to recover; but the plants in pots 4, 6, 23, and 24 became more wilted. Table 3 shows that these were pots in which nitrate concentrations in the drainage water collected on April 17 were high.

By April 27, avocado culture 1, which had temporarily recovered from the temperature peak of April 21, began to show severe wilt; plants in pots 2 and 3 showed considerable wilting; the plant in pot 22 showed increased wilt; and those in pots 4, 6, 23, and 24 had become progressively injured.

From this time until June 21, when the avocado series of cultures was discontinued, the affected plants became progressively worse, and some of them died. Examination of the roots as before showed extensive root rotting. All of the nonwaterlogged avocados continued healthy.

The grapefruit plants in the water-saturated soils, though showing no wilt at first, made no further top growth, and the lower leaves gradually began to show vein chlorosis, a condition which usually develops when serious root rotting occurs. Though the amount of vein chlorosis varied from plant to plant, there was no significant difference between the nitrate-treated and the nonnitrate-treated plants. On May 3, 1939, the greenhouse temperatures again reached a peak of nearly 100°F., and on the following day the leaves of the grapefruit plants in pots 21 and 22, receiving ammonium sulfate, began to curl. This is in harmony with the results of the first trial, in which the citrus plants first affected were those which had received ammonium sulfate. These two plants never fully recovered, though they remained alive.

As in the case of the avocados, the grapefruit cultures were continued until June 21, when the waterlogged plants were all so severely injured that they were discarded. As in the former experiment, no beneficial effects of nitrate under conditions of waterlogging were manifest, and those plants receiving ammonium sulfate were most affected.

The six nonwaterlogged grapefruit cultures of group B continued healthy except that the two plants receiving no nitrogen (pots 7 and 8) had become noticeably yellow from lack of nitrogen. It was decided to conduct a final experiment on these remaining six cultures to check again the effects of nitrate in the presence of excess water. The pots were leached prior to the addition of fertilizer, and nitrate determinations were made on the leachate. No trace of nitrate was found in any of the six pots.

Three of the pots, 8, 10, 11, were given $\text{Ca}(\text{NO}_3)_2$ in solution in amounts equivalent to a rate of 100 pounds per acre of nitrogen; the others were given the same volume of water containing no nitrate. The drainage holes of all six pots were plugged and the pots flooded, tar paper being placed over the top of the pots to prevent algal growth.

This experiment was continued until August 11, 1939. In harmony with former results, all of the waterlogged plants began to develop vein chlorotic leaves after about 10 days, the severity increasing with time. There was just as much vein chlorosis on the nitrate-treated plants as on those receiving no nitrogen. More or less of the curling which is characteristic of wilt took place. There was also marked mesophyll collapse in some of the leaves.

The nitrogen-starved plants in pots 7 and 8 were affected at about the same

rate and to about the same degree as those plants which were in a healthy green condition when waterlogging was begun. In general, the results of this last test were no different from those preceding.

DISCUSSION AND SUMMARY

The results of these experiments provide no evidence that additions of nitrate to waterlogged soils are beneficial. In fact, nitrate was definitely harmful to avocados. Ammonium sulfate was detrimental to both the avocados and the grapefruit.

The failure of additions of nitrate to produce beneficial effects in soil cultures deficient in oxygen is perhaps to be expected. Under favorable conditions of temperature and in the presence of suitable energy material, denitrification in most soils occurs rapidly. The analyses for nitrate made on drainage water from the pots on January 23 and again on February 27 indicated a rather rapid disappearance of this constituent. It is probable that under anaerobic conditions denitrifying organisms can make more rapid use of nitrate than can higher plants, since before it is available for respiration in the roots of higher plants nitrate must presumably be absorbed. Hoagland and Broyer (4), and Steward (5) have demonstrated the importance of an oxygen supply for nutrient accumulation. Hence, lack of oxygen would tend to prevent nitrate absorption, the first step requisite for the utilization of the combined oxygen in nitrate. Therefore, only such nitrate as the root contained at the time oxygen became limiting might be expected to be of value in compensating for oxygen deficiency.

The accentuation of waterlogging injury caused by nitrate in the case of the avocado and by ammonium sulfate in the case of both avocado and grapefruit seedlings may have been in part osmotic, in part a toxic effect from the production of nitrite or other products of nitrate reduction, and in part a stimulating effect on the growth of root-attacking organisms. Such a stimulation might produce a detrimental effect on the plant not only by increasing the number of organisms but by further accentuating oxygen deficiency.

Smaller additions of ammonium and nitrate might have produced less injury, and it is barely possible that lower concentrations of nitrate might have produced a favorable effect. Even the highest concentrations of nitrate as found by analyses of the drainage water are, however, well below the toxic limits of nitrate for these plants when grown under solution or sand-culture conditions.

Though it would be premature, on the basis of work with one soil and under only one set of conditions, to conclude that under practical conditions the addition or presence of ample nitrate will in no way offset the ill effects of oxygen deficiency, the results of these experiments offer no encouragement in this direction.

REFERENCES

- (1) ARNON, D. I. 1938 Ammonium and nitrate nitrogen nutrition of barley at different seasons in relation to hydrogen-ion concentration, manganese, copper, and oxygen supply. *Soil Sci.*, 44: 91-121.
- (2) CLEMENTS, F. E. 1921 Aeration and Air Content. The Role of Oxygen in Root Activity. Carnegie Institution of Washington, Washington, D. C.
- (3) HAAS, A. R. C. 1937 Nitrogen fertilization and root aeration. *Calif. Citrog.* 22: 286, 332, 333.
- (4) HOAGLAND, D. R., AND BROYER, T. C. 1936 General nature of the process of salt accumulation by roots with description of experimental methods. *Plant Physiol.* 11: 471-507.
- (5) STEWARD, F. C. 1935 Mineral nutrition of plants. *Ann. Rev. Biochem.* 4: 519-544.

PLATE 1

EFFECT OF AMMONIUM SULFATE AND CALCIUM NITRATE ON AVOCADO AND GRAPEFRUIT SEEDLINGS GROWING IN WATERLOGGED SOIL

A, No treatment; B, $(\text{NH}_4)_2\text{SO}_4$; C, $\text{Ca}(\text{NO}_3)_2$. The chemicals were applied in solution at the rate of 150 pounds of nitrogen per acre at the time the soils were waterlogged. The pictures were taken about 2 months later, all cultures having been kept saturated with water during this period.

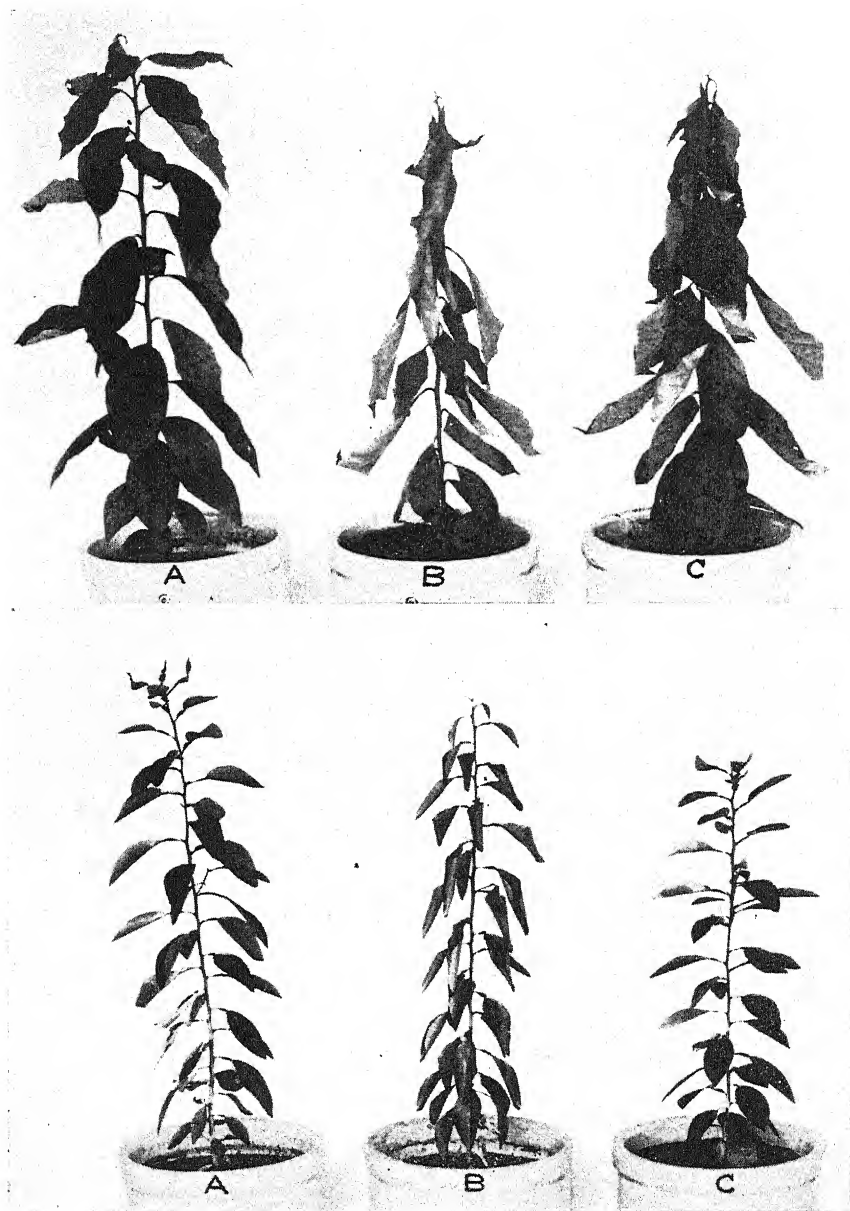
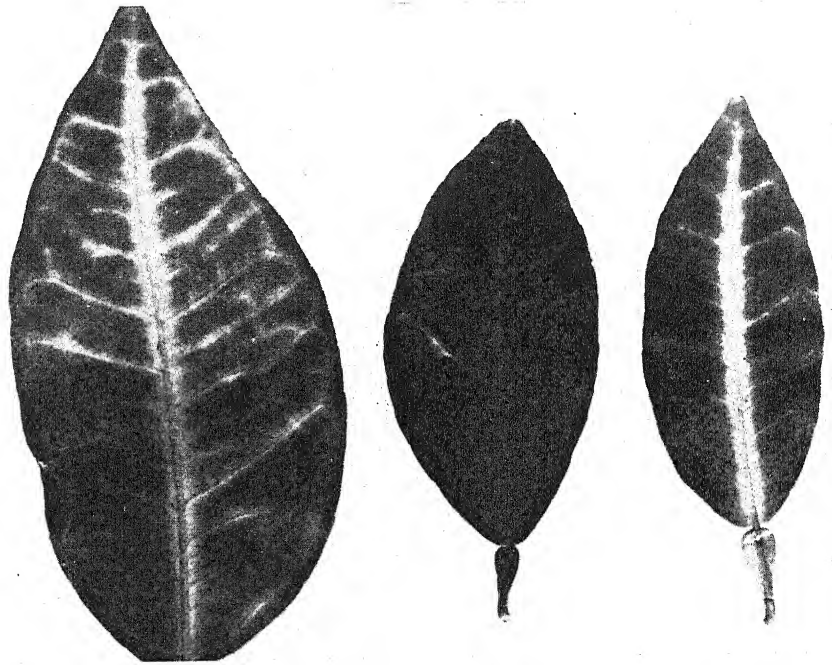


PLATE 2

VEIN CHLOROSIS IN CITRUS LEAVES RESULTING FROM ROOT ROTTING

This type of chlorosis may also be brought about by bark diseases as well as by mechanical injury to the conducting tissues of the roots, trunk, or branches. Even a break in the midrib of the leaf may cause yellowing of the veinal system beyond the break.



THE RELATION OF THE NUMBER OF TILLERS PER UNIT AREA TO THE YIELD OF WHEAT AND ITS BEARING ON FERTILIZING AND BREEDING THIS PLANT—THE SPACE FACTOR

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Plants require for their growth not only nitrogen, phosphorus, and other nutrients but also space for the expansion of their roots.² It is difficult to grow a tall plant in a small container, however favorable the other factors may be. In a field crop, as in a forest, a natural thinning takes place, and it is for this reason that the grain yield is, in large measure, independent of the quantity of seed sown or of the number of plants per acre.

Concerning natural thinning in field crops, we have shown in another paper (9) that the number of wheat tillers per unit area, or the "density" of the crop, increases up to a certain date and then decreases, only a small proportion of the tillers formed being recovered at harvest. The curve showing the variation in number of tillers or ears per unit area during growth, or the "density curve," and the number of tillers per unit area at the date of its maximum, or "maximum density," differ according to the variety. Under similar conditions, varieties that do not respond well to fertilization, as Katerini, attain a higher "maximum density" and have a sharper "density curve" and therefore a greater "relative density" than do varieties that respond well to fertilization. In other words, we have attributed the differences among varieties in response to nitrogen or phosphorus (9, 11) fertilizers, to differences in relative density, or the order in which the varieties are classed according to their maximum density.

This paper presents experimental data concerning density curves, maximum densities, and relative densities of wheat varieties and their effect on the response of the plant to fertilizers and the time of their application.

THEORETICAL CONSIDERATIONS

As has been shown elsewhere (10), no mathematical expression can accurately depict growth. Environment changes continuously during growth and must

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² For a full discussion of the space factor see *Ecologie Agricole* (10, chap. 7). The most plausible explanation of this need is that advanced by Pickering (12) that living roots excrete toxins.

be considered as comprising a series of instantaneous environments. Under the action of each instantaneous environment an instantaneous phenotype is produced, and a peculiar reactivity to environment corresponds to each phenotype.

Among the various mathematical expressions of growth, that of Balmukand (1),

$$\frac{1}{y} = \frac{a_n}{N} + \frac{a_p}{P} + \dots$$

fits the facts best. But we must consider this equation as expressing the influence of the environment on the growth of the plant during only one instantaneous environment, or at least a short period of development, and we must introduce into this equation the space factor.

A peculiarity of the space factor is that its value decreases as the crop grows. If X is the space available to the crop at sowing,³ the space available at the stage n of its development is $X - (fY_{n-1})$, fY_{n-1} being a function of the growth at the end of the preceding stage.

By introducing this factor in the Balmukand equation we have:

$$\frac{p_n(Y_{n-1})}{Y_n} = \frac{a_{nn}}{N_n} + \frac{a_{pn}}{P_n} + \dots + \frac{a_{xn}}{X - fY_{n-1}} \quad (A)$$

Where Y_n is the yield at the end of this stage; N_n , P_n , etc., the amounts of nitrogen, phosphorus, etc.; and a_{nn} , a_{pn} , etc., constants expressing the importance of each factor during this stage. In this equation we have replaced $\frac{1}{y}$ by $\frac{Y_{n-1}}{Y_n}$ because the growth at the end of a certain stage is a function of the growth at the beginning of the stage, and what is affected by environment is rather the ratio $\frac{Y_{n-1}}{Y_n}$ than Y_n ; p_n shows that the ratio depends also on the developmental phase. Equation (A) may be also written:

$$Y_n = \left[p_1 / \left(\frac{a_{n1}}{N_1} + \frac{a_{p1}}{P_1} + \dots + \frac{a_{x1}}{X} \right) \right] \left[p_2 / \left(\frac{a_{n2}}{N_2} + \frac{a_{p2}}{P_2} + \dots + \frac{a_{x2}}{X - fY_1} \right) \right] \dots \left[p_n / \left(\frac{a_{nn}}{N_n} + \frac{a_{pn}}{P_n} + \dots + \frac{a_{xn}}{X - fY_{n-1}} \right) \right] \quad (B)$$

The influence of each factor N , P , etc., on Y decreases with a decrease of any of the other factors. As the space available decreases during crop growth, the relative effectiveness of the available amounts of nutrients must decrease. In other words, fertile soils and heavy fertilization are more important when the crop is to be harvested at an early stage of development than when it is to be harvested at later stages.

Since grain is formed during the later stages of development, it is propor-

³ X from $\chi\acute{\omega}\rho\omicron\varsigma$ = space.

tionate rather to $Y_n - Y_{n-1}$, the growth during the later stage, than to Y_n , the total growth at the end of this stage. Equation (A) may be written:

$$Y_n - Y_{n-1} = Y_{n-1} \left[\left(p_n / \left[\frac{a_{nn}}{N_n} + \frac{a_{pn}}{P_n} + \dots + \frac{a_{zn}}{X - fY_{n-1}} \right] \right) - 1 \right] \quad (C)$$

to express the relation between grain yield and the growth at an earlier stage, that is, the maximum density. The relation is not linear. Y_n increases with Y_{n-1} up to a certain value of Y_{n-1} , for which

$$p / \left(\frac{a_{nn}}{N_n} + \frac{a_{pn}}{P_n} + \dots + \frac{a_{zn}}{X - fY_{n-1}} \right) = 1$$

Beyond this value of Y_{n-1} , Y_n decreases when Y_{n-1} increases. In other words, increments of yield of grain and straw corresponding to equal increments of maximum density gradually become smaller as a certain maximum density value is approached; beyond this value they become negative. This "critical" value of Y_{n-1} depends on N_n , P_n , etc., the quantities of nutrients available during the stage of grain formation. The greater the quantities of these nutrients or the more favorable the conditions, the higher is the critical maximum density. It is evident that for $Y_n - Y_{n-1}$ (grain yield) this critical density is sooner attained. From the foregoing relation between maximum density and yield we may conclude that high density varieties are adapted to poor soils or poor conditions and low density varieties to rich soils or favorable conditions. Likewise dense and early sowings are preferable in poor soils, and thinner and later sowings in rich soils.

Equation (C) may also be written:

$$\begin{aligned} Y_n - Y_{n-1} = & \left[p_1 / \left(\frac{a_{n1}}{N_1} + \frac{a_{p1}}{P_1} + \dots + \frac{a_{z1}}{X} \right) \right] \\ & \times \left[p_2 / \left(\frac{a_{n2}}{N_2} + \frac{a_{p2}}{P_2} + \dots + \frac{a_{z2}}{X - fY_1} \right) \right] \dots \\ & \dots \left[p_{n-1} / \left(\frac{a_{nn-1}}{N_{n-1}} + \frac{a_{pn-1}}{P_{n-1}} + \dots + \frac{a_{zn-1}}{X - fY_{n-2}} \right) \right] \\ & \times \left[\left(p_n / \left[\frac{a_{nn}}{N_n} + \frac{a_{pn}}{P_n} + \dots + \frac{a_{zn}}{X - fY_{n-1}} \right] \right) - 1 \right] \quad (D) \end{aligned}$$

to express the relation between grain yield and amounts of nutrients at different stages of development. Until the critical value of Y_{n-1} (maximum density) is reached, $Y_n - Y_{n-1}$ (grain yield) increases when N_1 increases, N_2 , N_3 , etc., remaining constant, and so on; in other words, the earlier the beginning of nutrient abundance or the later the beginning of nutrient starvation, the higher is the grain yield. But beyond the critical value of Y_{n-1} (maximum density), $Y_n - Y_{n-1}$ (grain yield) increases when N_1 , N_2 , N_3 , etc., decrease and N_n increases; in other words, early applications of nutrients are injurious, whereas very late applications (during the formation of the grain) are bene-

TABLE 1

Numbers of varieties and plots studied and mean maximum density and mean yield of each varietal type

FIELD	YEAR	LOCALITY	ECOLOGICAL REGION	S*	NUMBER OF VARIETIES†				NUMBER OF PLOTS‡				MAXIMUM DENSITY§				MEAN YIELD				OBSERVATIONS	
					M	X	R	Z	T	M	X	R	Z	M	X	R	Z	M	X	R		Z
1	'38	Vytina	High Mt.	N 26	39	9	1	75	118	176	52	13	359	190	215	240	290	430	510	550	810	Winter frosts
2	'38	Tripolitsa K.	Mediterr. N.	N 26	46	6	1	79	76	169	30	7	282	227	260	335	466	785	840	895	781	
3	'38	Larissa	Greece N.E.	E 24	47	2	..	73	60	170	8	..	238	243	257	318	...	865	930	822	
4	'39	Tripolitsa K.	Mediterr. N.	N 7	100	16	3	126	21	249	44	6	320	274	270	345	600	790	660	620	328	P. graminis
5	'39	Messara P.	Aegean	E 9	98	7	..	114	28	272	19	..	319	240	305	435	...	970	1,085	925	
6	'39	Thermi	Greece N.E.	N 14	109	10	..	133	38	294	28	..	360	250	305	415	...	920	1,135	1,360	
7	'38	Messara P.	Aegean	E 16	47	1	..	64	64	219	7	..	290	275	307	356	...	1,729	1,905	1,778	
8	'39	Messara F.	Aegean	L 11	99	9	..	119	30	269	20	..	319	275	307	356	...	1,729	1,905	1,778	
9	'39	Tripolitsa R.	Mediterr. N.	N 6	102	19	3	130	24	269	47	6	346	295	290	375	653	1,310	1,145	1,080	575	P. graminis
10	'38	Larissa	Greece N.E.	L 37	65	6	..	108	100	237	18	..	355	275	315	377	...	1,125	1,300	1,210	
11	'39	Dourouti	Mediterr. N.	N 5	84	18	3	110	24	265	59	10	358	300	300	350	520	795	665	600	370	
12	'39	Ptolemaida	Macedonia N.W.	N 6	31	41	13	91	27	127	124	37	315	303	335	430	570	2,505	2,290	2,520	2,240	
13	'38	Dourouti	Mediterr. N.	N 23	47	7	2	79	102	226	34	18	380	313	380	495	575	640	710	765	755	
14	'38	St. Mamas	Greece N.E.	L 7	24	1	..	32	13	77	5	..	95	320	378	370	...	2,090	2,045	2,570	P. graminis
15	'39	Vytina	High Mt.	N 7	31	41	12	91	27	136	121	36	320	317	386	450	655	1,095	1,070	1,110	970	
16	'38	Jannina	Mediterr. N.	N 23	18	4	1	46	90	82	31	10	213	353	392	437	632	2,430	2,510	2,520	1,965	
17	'38	Tripolitsa R.	Mediterr. N.	N 22	30	4	1	57	53	87	16	4	160	380	380	435	500	1,355	1,345	1,280	1,135	P. graminis
18	'38	Thermi	Greece N.E.	N 73	42	4	..	119	129	217	7	..	353	365	415	610	...	1,325	1,459	1,222	
19	'38	Messara F.	Aegean	L 16	51	1	..	68	54	207	8	..	269	357	434	514	...	980	1,105	550	
20	'39	Thessaloniki S.	Greece N.E.	S 20	34	2	..	56	88	144	8	..	240	390	425	460	...	795	825	840	P. graminis
21	'39	Thessaloniki S.	Greece N.E.	E 38	62	12	3	115	165	298	42	12	517	350	510	800	1,450	2,450	2,790	2,930	2,510	
22	'38	Thessaloniki D.	Greece N.E.	S 5	19	2	..	26	27	105	12	..	144	410	450	825	...	1,980	1,725	600	
23	'39	Larissa	Greece N.E.	L 4	81	8	..	93	22	246	19	..	287	375	495	600	...	1,890	1,810	1,385	
24	'38	Orestias	Greece N.E.	N 22	20	4	1	47	119	111	38	11	279	370	520	725	1,280	1,430	1,615	1,625	1,610	
25	'38	St. Mamas	Greece N.E.	E 8	24	1	..	33	35	119	5	..	159	418	474	558	...	3,140	3,130	3,240	

26	'38	Ptolemaida	Macedonia N.W.	N 31	5116	2100	148	226	79	20	473405	508	675	7683,090	2,865	3,070	3,040
27	'38	Thessaloniki S.	Greece N.E.	L 64	11119	4198	231	453	61	15	760422	500	592	7802,600	2,627	2,253	1,720
28	'39	Thessaloniki S.	Greece N.E.	L 38	6616	3123	155	304	46	12	517383	546	735	9202,240	2,360	2,090	1,720
29	'39	Orestias	Greece N.E.	N 11	326	..	49	46	25	..	210427	510	646	..	2,800	2,875	2,835
30	'39	Jannina	Mediterr. N.	N 13	297	150	37	77	21	4	139446	531	645	9682,665	2,405	2,180	2,120
31	'39	Larissa	Greece N.E.	E 4	798	..	91	22	246	20	288435	550	670	..	2,220	2,152	2,090
32	'38	Serres	Greece N.E.	E 42	628	..	112	118	220	26	364565	650	774	..	2,580	2,545	2,590
33	'39	Thessaloniki S.	Greece N.E.	N 45	6211	2120	175	191	27	6	399500	730	985	..	3,210	2,870	2,765
34	'38	Serres	Greece N.E.	L 41	649	..	114	115	215	26	356610	690	783	..	2,740	2,630	2,550
35	'38	Thessaloniki D.	Greece N.E.	E 61	9816	4179	252	472	66	19	809597	758	908	1,706	3,074	3,116	2,532
36	'39	Serres	Greece N.E.	L 42	6013	..	115	172	189	35	396840	1,008	1,155	..	4,990	4,255	3,760
37	'39	Serres	Greece N.E.	E 42	5912	..	113	175	191	33	399905	1,090	1,265	..	4,575	4,230	4,140
38	'38	Thessaloniki S.	Greece N.E.	N 43	6710	..	120	192	26	..	338885	1,215	1,565	..	3,970	3,673	3,441
Total.....														3,300	7,886	1,293	246
														12,725

* S = Sowing; E = early, N = normal, L = late, S = spring.

† M = varieties of density type Mentana, X = of density type Xylokastro, R = of density type Rieti, Z = of density type Zoulitsa, T = total.

‡ Plots each 10 (2 × 5) sq. m.

§ Density = number of tillers per square meter.

|| Yield = kilograms per hectare, multiplied by 1.121 gives pounds per acre.

ficial. We may conclude, therefore, that a moderate abundance of nutrients during the early stages of development and a very great abundance during the grain formation period are essential for a high grain yield.

EXPERIMENTAL

Density curves, maximum densities, and relative densities

In collecting data on density curves, maximum densities, and relative densities we have made frequent counts to determine the variation in the number of tillers per unit area of all our experimental fields in 1937-38 and 1938-39 (table 1). The number of these fields was 38, established in 12 experiment stations; the number of varieties, 553; the total number of fields \times varieties, 3558; and the total number of plots, more than twelve thousand.

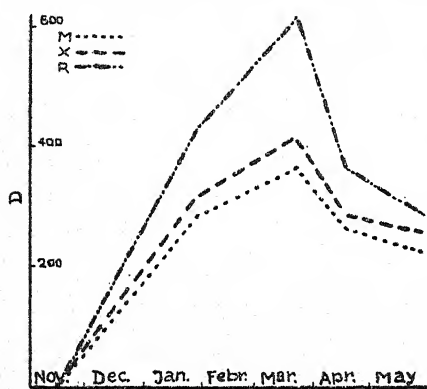


FIG. 1

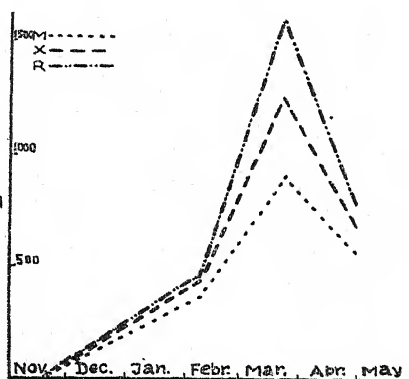


FIG. 2

FIG. 1. NUMBER OF TILLERS PER SQUARE METER AT DIFFERENT DATES OF THE VARIETIES OF THE DENSITY TYPES MENTANA (M), XYLOKASTRO (X), AND RIETI (R) ON THE VERY LOW FERTILITY FIELD AT THERMI IN 1938

FIG. 2. NUMBER OF TILLERS PER SQUARE METER AT DIFFERENT DATES OF THE VARIETIES OF THE DENSITY TYPES MENTANA (M), XYLOKASTRO (X), AND RIETI (R) ON THE VERY RICH FIELD OF THESSALONIKI SWAMP IN 1938

The relative density of varieties was the same in all experimental fields. The varieties may be classified, in the order of increasing density, into four types, as follows: Mentana, Xylokastro, Rieti, and Zoulitsa. The line of demarcation between any two types is only conventional, however, the varieties forming a continuous series of densities.

Density curves were plotted for each varietal type in each field. Curves for two of the fields are shown: the first (fig. 1), a very poor field (Thermi 1938); and the second (fig. 2), a very rich field (Institute swamp 1938) 2 miles away. It is to be noted that the density curves of all the varietal types are sharper in the fertile field than in the nonfertile; but the relative curvature for the different types is the same for the two fields, the sharpness of the curve being directly related to the density of the type. These observations apply also to the curves for the other 36 experimental fields.

Table 1 shows the maximum density of each varietal type in each experimental field. It is to be noted that the maximum densities of the types invariably follow the varietal classification already established.

TABLE 2

Mean relative densities, calculated from all 38 experimental fields, of certain varieties and their earing dates at Salonika and Ptolemaida

VARIETY	DENSITY	EARING DATES		VARIETY	DENSITY	EARING DATES	
		Salonika*	Ptolemaïda†			Salonika*	Ptolemaïda†
<i>Mentana type</i>				<i>Xylokaastro type</i>			
Fanfulla (Italian)....	422	Apr. 16	Asprostaro Peloponn. (Greek)....	664	Apr. 30	May 24
Hard White (Austrian).....	456	Deves (Greek).....	664	May 1
34288 (Greek).....	467	13	3130 (Greek).....	678	Apr. 24	15
Quaderna (Italian)...	469	17	18436 (Greek).....	685	22
31050 (Greek).....	481	14	May 9	Mavragani Kiprou (Cyprian).....	700	28
Kenya 10863 (Austrian).....	490	8567 (Greek).....	709	23
Rialle (Italian).....	492	13	Kenya 6041 (Austrian).....	718
31327 (Greek).....	493	13	Kenya 6040 (Austrian).....	724	26
Razza (Italian).....	502	15	<i>Rieti type</i>			
Kenya 6042 (Austrian).....	515	18	Psathas (Cyprian)...	777
Roma (Italian).....	527	20	29398 (Greek).....	778	Apr. 22	May 16
Mentana (Italian)...	531	13	Cologna (Italian)...	786	22
<i>Xylokaastro type</i>				Littorio.....	811
Eretria (Greek).....	583	Apr. 22	Ak Bassak (Greek)...	819	May 3
Kenya 10856 (Austrian).....	594	15	36095.....	850	Apr. 21	15
18372 (Greek).....	599	May 1	Rieti.....	860	May 1	24
Minos (Greek).....	614	Apr. 29	Klein 31C 9613 (Argentinian)....	860	Apr. 26
Limnos (Greek).....	617	27	May 16	38406 (Greek).....	875	13
Konduzi (Syrian)....	620	23	Prolifique (Yugoslavian).....	894	May 5	26
33497 (Greek).....	632	15	38414 (Greek).....	895	17
Santa Maria (Italian)	640	May 1	Terminillo (Italian)	921	Apr. 24	25
Ayros (Greek).....	646	Apr. 23	Chondrostaro (Greek).....	981	May 6
Mavragani Vytinis (Greek).....	653	May 10	May 23	<i>Zoulitsa type</i>			
Xylokaastro (Greek)...	653	Apr. 26	38142 (Greek).....	1150	May 12
Canberra (Australian).....	660	Katranitsa (Greek)	1237	May 5	22
				Zoulitsa.....	1514	5	29

* Sowing date, November 3, 1938.

† Sowing date, October 28, 1938.

Table 2 shows the mean relative densities, for all the experimental fields, of a limited number of varieties and their dates of earing at Salonika and at

Ptolemaïda. It is evident that varieties of low density are generally early, and those of high densities, late. Earliness affects density because early varieties have not the time for profuse tillering, but the characteristic density of a variety does not depend solely on its periodicity.

Relation of maximum density to grain yield

On the basis of the aforementioned data, curves were drawn showing the relation of maximum density to yield for each type in each experimental field.

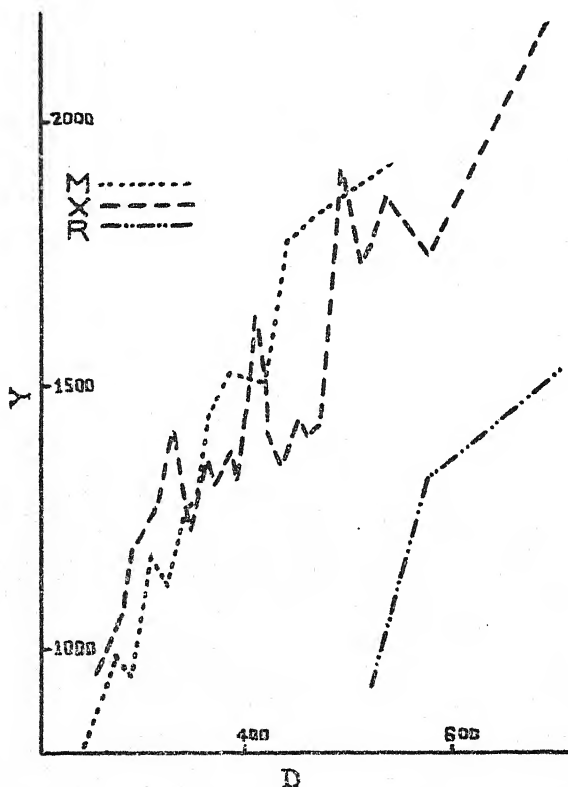


FIG. 3. RELATION BETWEEN YIELD (Y) AND MAXIMUM DENSITY (D) OF THE VARIETIES OF THE DENSITY TYPES MENTANA (M), XYLOKASTRO (X), AND RIETH (R) ON THE THERMI FIELD IN 1938

Each point of the curves represents 10 plots

Figures 3 and 4 show the curves of the two fields for which the density curves have been given (figs. 1, 2).

The curves fully confirm the theoretical conclusions already reached, that to equal density increments there are corresponding yield increments which become smaller as the critical density is approached; beyond this point, no

yield increments correspond to density increments. Furthermore, this critical density, which is lower for poor than for rich fields, is attained sooner in the latter fields (fig. 4) than in the former (fig. 3) with varieties of higher density.

Relation of the relative densities of varieties to their adaptation to different fertility levels

As high density varieties attain their maximum densities more readily than do low density varieties, their yields are less influenced by soil fertility differences. In consequence, they are better adapted to poor soils or poor conditions. This fact is illustrated by the following mean yields (kilograms per

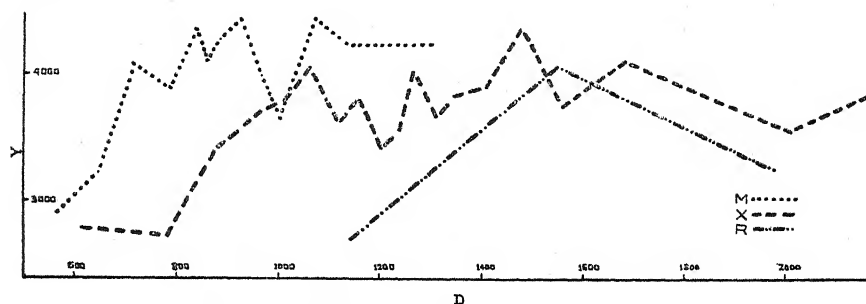


FIG. 4. RELATION BETWEEN YIELD (Y) AND MAXIMUM DENSITY (D) OF THE VARIETIES OF THE DENSITY TYPES MENTANA (M), XYLOKASTRO (X), AND RIETI (R) ON THE FIELD OF THESSALONIKI SWAMP IN 1938

Each point of the curves represents 10 plots

hectare⁴) of the different density types in four fields arranged according to their fertility:

DENSITY TYPE	VYTINA, 1938 (VERY POOR)	THERMI, 1938 (POOR)	THESSALONIKI SWAMP, 1938 (RICH)	SERRES LATE, 1939 (VERY RICH)
Mentana.....	430	1325	3970	4990
Xylokastro.....	510	1459	3673	4255
Rieti.....	550	1222	3441	3760
Zoulitsa.....	810

The highest yield was produced in the very poor field of Vytina by the varieties of the highest density (type Zoulitsa); in the poor field of Thermi, by the varieties of the less dense type Xylokastro; and in the rich fields of Thessaloniki swamp soil and Serres, by the varieties of the lowest density (type Mentana). Thus the theoretical conclusions have been fully confirmed.

⁴ One kilogram per hectare corresponds to 1.121 pounds per acre.

The differential response of varieties to fertilization and its relation to their relative density

Field experiment with nitrogen at Thermi in 1937. Four varieties, Deves, 3130, Eretria, and Mentana, were sown on the poor field at Thermi with and without nitrogen fertilizers, applied as follows: 150 kgm. of ammonium sulfate (21 per cent N) per hectare at sowing and 250 kgm. of sodium nitrate (16 per cent N) on March 12. Superphosphate (16 per cent P_2O_5) was applied with the ammonium sulfate to all plots at the rate of 187.5 kgm. per hectare. The following yields (kilograms per hectare) were obtained:

VARIETY	WITHOUT NITROGEN	WITH NITROGEN	INCREMENT
Deves.....	900	1150	250
3130.....	1180	1660	480
Eretria.....	1000	1600	600
Mentana.....	700	1510	810

Number of replicates, 5; error of the means, 90.8.

The increment was about twice as great for the varieties of low density (Mentana and Eretria) as for those of high density (Deves and 3130). Without nitrogen the varieties of higher density and with nitrogen those of lower density yielded better.

Field experiment with phosphorus at Thermi in 1938. Two varieties, Deves and Mentana, were sown, also on the poor field of Thermi, with and without phosphorus fertilizers. The quantities of fertilizers applied were 200 kgm. per hectare of ammonium sulfate to all plots and 376 kgm. of superphosphate to half the plots. The yields (kilograms per hectare) were as follows:

VARIETY	WITHOUT PHOSPHORUS	WITH PHOSPHORUS	INCREMENT
Deves.....	1150	1480	330
Mentana.....	1045	1757	712

Number of replicates, 8; error of the means, 106 kgm.

The results are similar to those with nitrogen on this field in 1937. The density curves of this experiment agree closely with those of figure 5, obtained with phosphorus fertilization in pots.

Field experiment with nitrogen at Thessaloniki in 1938. The same two varieties, Mentana and Deves, were sown on the rich field at Thessaloniki with and without nitrogen fertilizers, applied as follows: 300 kgm. per hectare of ammonium sulfate at sowing and 160 kgm. sodium nitrate on February 4 and again on April 4. The following yields (kilograms per hectare) were obtained:

VARIETY	WITHOUT NITROGEN	WITH NITROGEN	INCREMENT
Deves.....	1376	2353	977
Mentana.....	1338	2640	1302

Number of replicates, 10; error of the means, 94.5 kgm.

The results are similar to those on the poor field in 1937, and the density curves again agree closely with those of figure 5.

Field experiment with nitrogen, phosphorus, and potassium at Thessaloniki in 1937-38. In 1937-38, 85 varieties (25 of the Mentana density type, and 60 of the Xylokastro) were sown with and without complete fertilizers, each variety in five pairs of plots. The fertilizers applied per hectare were 300 kgm. of ammonium sulfate, 400 kgm. of superphosphate, and 140 kgm. of potassium

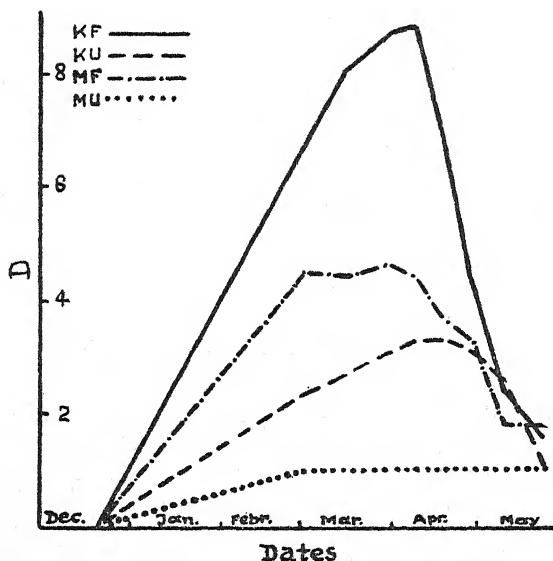


FIG. 5. NUMBER OF TILLERS PER POT AT DIFFERENT DATES OF THE VARIETIES MENTANA (M) AND KATERINI (K), UNFERTILIZED (U) AND FERTILIZED (F)
—PHOSPHORUS SOIL EXPERIMENT, 1939

chloride at sowing; 260 kgm. of sodium nitrate on February 4 and again on April 4. The mean yields (kilograms per hectare) were as follows:

DENSITY TYPE	UNFERTILIZED	FERTILIZED	INCREMENT
Mentana.....	1738	3684	1946
Xylokastro.....	1810	3552	1742

The results are similar to those in the preceding experiments.

Field experiment with nitrogen and phosphorus at Thessaloniki in 1939. In 1939, thirty-two varieties (13 of the Mentana type of density, 18 of the Xylokastro, and 1 of the Rieti) were sown with and without nitrogen and phosphorus fertilizers, each variety in five pairs of plots. The fertilizers applied per hectare were 600 kgm. of ammonium sulfate and 600 kgm. of superphos-

phate at sowing, and 200 kgm. of sodium nitrate on February 13 and again on March 3 and 23. The mean yields (kilograms per hectare) were as follows:

DENSITY TYPE	UNFERTILIZED	FERTILIZED	INCREMENT
Mentana.....	2454	3499	1045
Xylokastro.....	2508	3279	771
Rieti.....	2929	3694	765

The results do not differ from those already reported; again the density curves agree closely with those of figure 5; and curves depicting the relation between maximum density and grain yield for every group of varieties, unfertilized and fertilized, agree with those of figure 3 and 4 (poor and rich fields).

The theoretical conclusions have thus been fully confirmed by all these field experiments.

Response of wheat to different dates of fertilization

In studying the response of wheat to different dates of fertilization we have used the procedure described by the author in another paper (11). Two wheat varieties, one of low density (Mentana) and the other of high density (Katerini), were each sown in two series of pots, one maintained at a constant high level of the nutrient considered and the other maintained at a constant low level. At short intervals one pot from each series was transferred to the other, that is, from starvation to abundance and *vice versa*.

A constant high level of phosphorus was maintained by sowing the plants in small pots containing 200 gm. of soil low in phosphorus and placing under the pots glasses containing 700 cc. of a solution of 1 per cent NaH_2PO_4 and 1 per cent $\text{CaH}_4(\text{PO}_4)_2$. For a constant low level of phosphorus, the glasses contained only water.

A constant high level of nitrogen was maintained in the pots in 1937 by weekly leaching with a 4 per cent solution of calcium nitrate, and in 1938 with 1 per cent calcium nitrate and 1 per cent sodium nitrate solution, the volume of solution used being about four times as great as the maximum water-holding capacity of the soil. A constant low level of nitrogen was maintained in the pots by leaching at the same intervals with water. In 1938-39, the procedure followed with the phosphorus was applied to the nitrogen experiment.

Curves showing the relation between grain yields or maximum densities and dates of transfer from starvation to abundance and *vice versa* were plotted. Table 3 gives characteristic figures of the curves.

In general, the relation between the date of transfer from starvation to abundance of a nutrient (nitrogen or phosphorus) and maximum density is a simple one: the earlier the transfer, the higher is the density. But the relation is not linear; as the transfer is delayed, the number of tillers falls very slowly until about mid-March (40 days before earing), after which it falls very rapidly.

The relation between the date of transfer from starvation to abundance of

TABLE 3

Maximum densities (tillers per pot) and grain yields (grams per pot) of the wheat varieties Mentana and Katerini when transferred from starvation to abundance or vice versa at certain characteristic dates, between which the variation is approximately linear

FROM STARVATION TO ABUNDANCE						FROM ABUNDANCE TO STARVATION					
Mentana			Katerini			Mentana			Katerini		
Date	Maxi- mum den- sity	Yield	Date	Maxi- mum den- sity	Yield	Date	Maxi- mum den- sity	Yield	Date	Maxi- mum den- sity	Yield
<i>Nitrogen experiment of 1937</i>											
Dec. 20	11.8	14.8	Dec. 20	18.9	9.72	Dec. 20	8.8	11.1	Dec. 20	12.2	10.52
Mar. 28	12.2	14.0	Mar. 10	18.5	7.50	May 16	14.0	14.8	Apr. 18	21.2	9.72
June 8	8.8	11.1	June 8	12.2	10.52	June 8	11.8		June 8	18.9	
<i>Nitrogen experiment of 1938</i>											
Mar. 20	6.4	3.7	Mar. 20	12.2	4.70	Mar. 20	7.0	3.5	Mar. 20	12.2	3.30
May 6	3.5	1.7	May 6	6.0	2.30	Apr. 16	11.0	9.2	Apr. 11	16.2	7.20
						Apr. 26	10.0		May 4	17.2	
<i>Nitrogen solution experiment of 1939</i>											
Jan. 14	6.5	1.70	Feb. 20	8.5	1.00	Jan. 14	2.8	0.95	Feb. 20	8.8	0.54
Mar. 15	5.0	1.05	Apr. 1	7.7	0.70	June 2	6.5	1.80	Apr. 12	14.0	1.00
May 16	1.5	1.52	May 16	3.8	1.09	Apr. 20	6.5	1.80	May 23	12.0	1.00
		0.80	May 16		0.80	May 16					
<i>Phosphorus solution experiment of 1939</i>											
Jan. 19	3.7	1.77	Feb. 2	6.0	0.77	Jan. 26	3.1	1.33	Feb. 20	7.9	0.85
Apr. 14	3.2	1.30	Mar. 10	4.2	0.83	Mar. 14	4.5	2.20	Apr. 20	9.6	1.40
May 26	1.6	1.67	May 12	4.0	0.40	May 30	4.5	1.67	May 30	8.5	1.40
		0.68	May 26								

Date of heading about May 1 (earlier for Mentana, later for Katerini).

the nutrient (nitrogen or phosphorus) and grain yield is more complex. When the need for the nutrient is pronounced, as in the nitrogen experiment of 1938, the relation is linear for both varieties: the earlier the transfer, the higher is the yield. But when the need for the nutrient is less pronounced, as in the nitrogen experiment of 1939, there are two points of inflection; the yield falls until the first date, after which it rises until the later point of inflection, which corresponds roughly to heading date, and then falls rapidly. When the need for the nutrient is still less, as in the nitrogen experiment of 1937, the abundance of the nutrient is injurious to the grain yield of the high density variety when it begins at the earlier point of inflection.

The relation between the date of transfer from abundance to starvation and maximum density or grain yield is simple: the earlier the transfer, the lower is the density or the yield in general, but beyond a certain date neither the density nor the yield is appreciably influenced.

The foregoing results are in full agreement with the theoretical conclusions already reported. As has been pointed out, up to a certain value of maximum density, the critical density, the influence of the abundance on grain yield is greater the earlier it begins; in other words, the curve is continually falling. Beyond this density, early abundance is injurious and only late abundance (about at heading) is favorable; in other words, the curve presents a maximum about at heading. The combination of these two curves gives, for intermediate conditions, a curve with two points of inflection, an elevation about at heading and a fall before heading.

It is to be noted that the transfer from a constant low level of a nutrient to a constant high level at a series of dates does not correspond to applications of fertilizers at these dates. In an unfertilized soil the amount of nitrogen is not constant (10), and fertilizations do not raise this level to a constant higher one, the nitrogen or phosphorus added being partly immobilized.

The experiments of Richardson (13) in Australia, of Yankovitch (15) in Tunisia, and of Tulaikov (14) in Russia showed that the amount of nitrate nitrogen in the soil of a small-grain field decreases from sowing to harvest, and is at the minimum during the period of grain formation. As a high level of nitrogen is essential during this period of growth, we may conclude that a late application (a little before heading) of nitrogen is always favorable. If the soil is poor, an early application is also necessary to raise the maximum density to each critical value.

The fact that the action of phosphate fertilizers does not last long suggests the idea that the amount applied at sowing is also partly immobilized until harvest. If this is so, the foregoing conclusions concerning nitrogen fertilizers are also applicable to phosphorus fertilization. In order to elucidate this question the following experiment was carried out in 1939:

Pots containing 850 gm. of a low-phosphorus soil fertilized with nitrogen and sown with Mentana or Katerini were fertilized with 0.3 gm. of superphosphate on various dates. Another series of pots received 0.15 gm. at sowing and 0.15 gm. on various dates. Table 4 shows characteristic figures of the

TABLE 4

Maximum density (tillers per pot) and grain yield (grams per pot) of the wheat varieties Mentana and Katerini when fertilized with super phosphate at certain characteristic dates, between which the variation is approximately linear

HALF AT SOWING AND THE REMAINDER AT DIFFERENT DATES									
SINGLE APPLICATION									
Mentana					Katerini				
Date	Maxi- mum den- sity	Date	Yield		Date	Maxi- mum den- sity	Date	Yield	
Dec. 12	4.6	Dec. 12	1.56		Dec. 18	9.0	Dec. 18	1.16	
Mar. 14	3.8	Mar. 4	1.40		June 6	3.0	Apr. 10	1.36	
May 1	1.0	June 6	0.44		June 6		June 6	0.84	
June 6	1.0								

curves obtained. The need for phosphorus was pronounced; 0.3 gm. of superphosphate increased the yield of Mentana 229 per cent and that of Katerini 144 per cent. The earlier the single application of phosphate was made, the higher were the maximum density and the yield. The relation is not linear: up to about the end of March (30 days before earing), maximum density and yield decrease relatively slowly when the application is delayed; after this date, the decrease is more rapid. When the phosphate was applied in two doses, the second dose did not appreciably affect maximum density, but it did increase grain yield, especially when applied about April 8 (20 days before heading). We may conclude from these results that, for a high grain yield, an abundance of available phosphorus during grain formation is essential, and this cannot always be obtained by the application of mineral phosphorus fertilizers at sowing.

In all these experiments, as in the field experiments, the low density variety, Mentana, responded better to fertilization than did the high density variety, Katerini. The yields (grams per pot) were as follows:

EXPERIMENT	UNFERTILIZED		FERTILIZED		INCREMENT	
	Mentana	Katerini	Mentana	Katerini	Mentana	Katerini
Nitrogen 1937.....	11.10	10.52	14.80	9.72	3.70	-0.80
Nitrogen 1938.....	1.70	2.30	9.20	7.20	7.50	4.90
Nitrogen 1939.....	0.80	0.54	1.80	1.00	1.00	0.46
Phosphorus (solution) 1939.....	0.68	0.40	1.68	1.40	1.00	1.00
Phosphorus (soil) 1939.....	0.44	0.40	1.56	1.16	1.12	0.76

The respective density curves all agree closely with one another and with those found by the author (11) in previous experiments. Figure 5 shows the curves of one experiment. In short, the results of these pot experiments with nitrogen and phosphorus fully confirmed the theoretical conclusions.

The space factor

Natural selection. In a previous paper (7) we showed that natural selection among wheat varieties sown mixed does not always favor the varieties that give the higher grain yields when sown alone under the same conditions, and we concluded that (a) high grain yield does not constitute the only factor on which depends dominance in the struggle for existence; (b) wheat varieties differ in aggressiveness, and their classification in this respect is remarkably constant.

It is to be noted that the more aggressive varieties, as Canberra, Mykine, and Xylokastro, are of higher maximum density, and the less aggressive varieties, as Mentana, are of lower maximum density. It is conceivable that when two varieties of different maximum density are sown together, the one forming the more numerous tillers will choke the other. Curves of density would appear, therefore, to be very valuable in the study of natural selection.

On the premise that varieties of high density are poor soil varieties and that

natural selection favors varieties of high density, it must be concluded that natural selection chooses genotypes adapted to poor environmental conditions.

Pot experiments on varieties. Previous papers (4, 5) described the pot method used by the author for the study of wheat varieties. When the pots are small in comparison with the number of plants grown in them, the space factor assumes primary importance, and varieties adapted to fertile soil are favored. Indeed, there are always low density genotypes (of the Mentana type) which are selected on the basis of pot experiment yields. For this reason, we began in 1937 to use very small containers (800 gm. of soil), heavily fertilized, and in which only one wheat plant is grown, for the selection of progeny adapted to fertile soils or abundant fertilization. On the other hand, we use large containers (30 kgm.) filled with poor soil, and bearing only one plant, for the selection of genotypes adapted to poor conditions.

Pocket experiments on varieties. The pocket method has been described in detail in previous papers (6, 8). Briefly, in this method wheat plants are widely spaced (20 by 20 inches); the pockets of each strain are arranged in several lines; the pockets of each line are sown with two strains; and the pairs of strains sown in the same line are different for each repetition. By this method, genotypes adapted to poor soil and resistant to *Puccinia graminis*⁵ are selected.

Organic fertility requirements. It is well known that the high yields of wheat in fertile soils cannot readily be obtained with abundant fertilization in poor soils. According to Damseaux (2) old fertility (*vieille force*) is required for these higher yields. This fact can now be explained by the space factor. When abundant mineral fertilization is applied at sowing, all the nitrogen becomes available at once to the crop, and a great deal of it is lost or immobilized before heading. Dense growth results, and this depresses grain yield. On the other hand, in a fertile soil, the nitrogen becomes available to the crop in small fractions which do not induce too high density and which ensure a high level of available nitrogen during grain formation. By applying the large quantities of mineral fertilizers in many fractions, the last late in the growing season, we can obtain high yields with mineral fertilizers in poor soils.

Experiments on fertilizer requirements of soil. When the fertilizer requirements of soils are studied by means of field or pot experiments, the plants are usually harvested at maturity, and the results are thus complicated by the space factor. It would be better to harvest the plants at an early stage or merely to count the number of tillers per unit area. This method is simpler, cheaper, and more rapid, and the differences between fertilized and unfertilized soils are augmented, because, as has been shown, the effectiveness of fertilizers is greater the earlier the crop is harvested.

⁵ *Puccinia graminis* severely attacks wheat plants grown at great distances from one another and profusely tilled.

DISCUSSION

Though the experiments reported in this paper deal only with wheat, it is evident that analogous facts could be observed in other plants. Thinning and pruning of trees, for instance, could also be studied from the point of view of the space factor.

SUMMARY

Numerous facts show that plants require for their growth not only nitrogen, phosphorus, and other nutrients, but also space for the expansion of their roots. By introducing this factor into Balmukand's equation of growth, as modified by the author, equations have been developed which show the following facts: Fertile soils and heavy fertilizations are more important when the crop is to be harvested at an early stage than at later stages. In general, there is a positive correlation between growth density at an early stage and grain yield, but this relation is not linear, for increments of grain yield corresponding to equal increments of density gradually become smaller until a certain critical density is reached, beyond which the yield does not increase, or even decreases as density increases. This critical density is higher as the conditions during grain formation are more favorable. In consequence, a too dense early growth depresses grain yield. Varieties with dense early growth cannot attain the high grain yields of varieties with less dense early growth; the first are better adapted to poor soils or poor conditions. A high grain yield necessitates favorable conditions during early growth, but not so favorable as to induce a density exceeding the critical one; it also necessitates very favorable conditions during grain formation.

Frequent counts were made during 2 years on the number of tillers per unit area of all the wheat varieties on more than twelve thousand plots on the experimental fields at 12 different stations of the Greek Plant Breeding Institute. The number of tillers increases to a certain date and then decreases. The maximum density depends not only on environmental conditions but also on variety. The order in which the varieties are classed with respect to their maximum density or their relative density was approximately the same in all the 38 experimental fields. In general, early varieties are low density varieties, but there are varieties which combine earliness with high density.

A comparison of the maximum density of each plot with its grain yield confirmed the theoretical conclusions concerning the relation of early growth to grain yield. Varieties of high density yield better on poor soils, varieties of low density on rich soils; the latter may attain higher grain yields.

Special field experiments were carried out on the differential response of varieties to nitrogen, to phosphorus, and to both. In all these experiments the grain yield increments were considerably greater for low density than for high density varieties.

Pot experiments in which wheat plants were transferred on different dates from starvation to abundance of nitrogen or phosphorus and *vice versa* also

confirmed the theoretical conclusions that a high grain yield necessitates a moderately high level of fertility during early growth and a very high level during grain formation. In all these experiments also, the variety of low density responded better than that of high density to an abundance of nutrients. On the premise that the amount of nitrogen in an unfertilized soil decreases from sowing to maturity and that added mineral fertilizers are sooner or later immobilized, it must be concluded that fertilizers must be applied early only on poor soils, and then not too profusely, and natural fertility or early application must be supplemented by an application sufficiently late to supply an abundance of nutrients during grain formation. This was confirmed by pot experiments in which phosphorus was added in single applications on different dates and in fractional applications, one at sowing and the other on different dates.

The order in which the varieties are classed according to their density is similar to that of their aggressiveness. One may conclude, therefore, that high density of early growth is one of the main factors of domination in the struggle for existence, and that natural selection chooses genotypes adapted to poor environmental conditions.

The author's pot method employing small containers of heavily fertilized soil is very valuable for the selection of genotypes adapted to rich soil; and that employing large pots of poor soil, for the selection of genotypes adapted to poor soil. The pocket method, in which varieties are widely spaced, also is valuable for the selection of genotypes adapted to poor soil.

The experiments explain why the high yields obtained on fertile soils cannot readily be obtained with single large applications of mineral fertilizers on poor soils and why fractional applications give better results.

In experiments on fertilizer requirements of soils, the results are complicated by the space factor when plants are harvested at maturity. Harvesting at an early stage would be better and more convenient, or when a small cereal plant is used, it should be sufficient to count the number of tillers per unit area.

REFERENCES

- (1) BALMUKAND, B. 1928 Studies in crop variation: V. The relation between yield and soil nutrients. *Jour. Agr. Sci.* 18: 602-627.
- (2) DAMSEAUX, A. 1911 Plantes de la Grande Culture.
- (3) LYON, T. L., BIZZELL, J. A., AND WILSON, B. D. 1923 Depressive influence of certain higher plants on the accumulation of nitrates in soil. *Jour. Amer. Soc. Agron.* 15: 457-467.
- (4) PAPADAKIS, J. S. 1933 Method of evaluation of drought resistance. Thessaloniki Plant Breeding Inst. Sci. Bul. 13.
- (5) PAPADAKIS, J. S. 1935 Varieties experiment in pots. Thessaloniki Plant Breeding Inst. Sci. Bul. 20.
- (6) PAPADAKIS, J. S. 1935 The pocket method of varieties experiments. Thessaloniki Plant Breeding Inst. Sci. Bul. 21.
- (7) PAPADAKIS, J. S. 1937 Est-ce seulement d'après le rendement en grain que se fait la selection naturelle chez les plantes cultivées? Thessaloniki Inst. Amélior. Plantes Bul. Sci. 26.

- (8) PAPADAKIS, J. S. 1937 Expériences et perfectionnements à la methode des pockets pour des essais de variétés. Thessaloniki Inst. Amelior. Plantes Bul. Sci. 27.
- (9) PAPADAKIS, J. S. 1938 Expériences sur la localisation des engrais et la fertilisation phosphorique du blé. Thessaloniki Amelior. Plantes Bul. Sci. 31.
- (10) PAPADAKIS, J. S. 1938 Ecologie Agricole.
- (11) PAPADAKIS, J. S., AND TALLELIS, D. 1937 Expériences sur la fertilisation azotée du blé et la différenciation des variétés par rapport à l'azote. Thessaloniki Inst. Amelior. Plantes Bul. Sci. 30.
- (12) PICKERING, S. U., AND BEDFORD, DUKE OF. 1903 Reports of the Woburn Experimental Fruit Farm. London.
- (13) RICHARDSON, A. E., AND FRICKE, E. F. 1931 Effect of nitrogenous fertilizers on the growth and yield of wheat and barley. *Jour. Dept. Agr. S. Aust.* 35: 57-86.
- (14) TULAIKOV, N. M. 1937 Fractional application of nitrogenous fertilizers on spring wheat under irrigation. *Soil Sci.* 44: 293-298.
- (15) YANKOVITCH, L. 1933 Le problème de l'azote dans les terres Nord Africaines. *Ann. Serv. Bot. [Tunis]* 10.

STRUCTURAL CHARACTERISTICS OF PEAT AND MUCK

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The morphological study of peat areas is a relatively recent line of investigation. The probable reasons for this fact are as follows: areas of peat as a subject of scientific research differ in origin and development from areas of mineral soils; the nature and morphological features of the main regional types of peat deposits have not yet been clearly ascertained; relationships between structural characteristics and the composition, properties, and environmental conditions of different types of peat and muck are far from well understood; and areas of peat and muck are but very feebly altered by soil-making processes leading to the development of eluvial and illuvial horizons. Unlike mineral soils, which are a physical and chemical laboratory and the home of countless, living microorganisms, earthworms, and other kinds of life, where leaching, translocation, and reactions of the greatest importance are constantly occurring, peat deposits in their natural condition are relatively lifeless and inert. It is only at the surface, where the vegetative cover is anchored, that water and very small amounts of nutrients are absorbed by a variety of root systems and underground parts of plants, and that activities are carried on which contribute to a continuous accumulation of plant remains on land and in open water.

STRATIGRAPHIC FEATURES OF PEAT AREAS

Among the several lines of inquiry that may be applied to deposits of peat as a subject of scientific research is the study of stratigraphic, comparative, and developmental morphology. It is not generally recognized that one of the most distinctive features of an area of peat is the sequence or arrangement of layers below the surface. Stratification arises from differences in color, texture, and composition of plant remains that accumulate under conditions of changing environment. The examination and comparison of plant remains in layers of peat from successive stages of peat-forming vegetation help to give an understanding of the epitomized history of external or environmental conditions expressed in positive terms. Hence the characteristics and habits of the peat-forming vegetation, the influences of climate and topography, and the modifying conditions of ground water and its content of soluble salts are the more important factors which are instrumental in the development of a peat area and which determine the character of profile differ-

ences and the classification of peat land. In some cases the stratigraphic similarities are so clear that the degree of unity and constancy in morphological profile features with respect to the sequence of peat layers and related succession of vegetation and in the reconstruction of environmental conditions cannot be doubted. They afford knowledge which may be held as positive and useful in the classification and valuation of peat land. It is from this source, also, that age determination, correlations, and the conclusions of comparison between American and Eurasian peat deposits may often be effectively tested (3). But in other cases the origin and stratigraphic features of areas of peat are not well known. Probable causes of differences in individual profile characteristics appear to be changes in the composition of successions of vegetation, in water table, and in supply of mineral nutrients; they may be due to seasonal changes in rate of accumulation of plant remains and organic mixtures which give rise to "varved" sediments, gyttja, and dy, or they may be caused by drought, floods, erosional cycles, and even changes in sea level and volcanic eruptions.

Fundamental types of peat profiles and peat materials have been illustrated and described by the author (2). Other pertinent contributions include: data in the form of descriptions illustrating the characteristics of peat profiles of certain areas in Pacific Coast States (6) and of various North and South American peat deposits (5); analytical data on different kinds of peat material (1, 4). The physical and chemical properties of entire peat profiles from various sections of the United States have been presented by Feustel and Byers (7). Adequate field descriptions and coordinated laboratory tests of samples from the entire depth of a peat area are of distinct theoretical significance and great practical importance; they constitute the best basis for correct conclusions as to the main processes which give rise to differences in stratification and to the main properties of the various kinds of plant remains designated by the term "peat" as well as their products of decomposition designated by the term "muck."

Peat- and muck-forming processes have certain characteristics in common with soil-forming processes: diverse kinds of peat and muck develop because of differences not only in vegetation and the geographical landscape, but also in the relative intensity of such processes as "leaching and evaporation, decomposition, synthesis, solution and precipitation, oxidation and reduction, hydrolysis and neutralization." The results, however, are not the same as in mineral soils. Many of the processes have been cyclic in nature, warm periods following cold periods, wet periods following dry periods. These cyclic processes have resulted in changes which the morphologist can understand, if he keeps in mind the long-time lag between the conditions and the related changes in the development of a certain type of peat profile. Profile characteristics endure long after the forces responsible for their development have disappeared. This fact may aid in settling some of the disputes between morphologists, chemists, soil scientists, and others interested in the problem

of morphological features of peat land and mineral soils or in the study of peat resources and their uses.

STRUCTURAL FORMS OF PEAT MATERIAL

Every area of peat and every profile section within the areal limits of a peat deposit exhibit not only their own characteristic stratification or arrangement of layers but also visible differences in size and grouping or spacing of the components and aggregates of which layers of peat are composed. Definite types of structural arrangement in primary peat materials are closely connected with the composition, size, shape, and stability of the components and aggregates. Decomposition and cultivation exercise a destructive effect on the character, color, consistence, and condition of the original peat material. Depending on the moisture condition, the material may be in a loose, friable state of consistence, completely granulated, or it may finally culminate in a more or less colloidal sticky form of organic residue that is hard and structureless when in a dry condition.

The structural form of peat materials is indeed a basis of classification but in itself it is essentially limited in application. This is shown by the fact that conclusions drawn from it have to be constantly checked by the examination of other characteristics and particularly those relating to botanical composition.

Examination of peat materials by means of the microscope for purposes of comparison has made very rapid strides in recent decades. The comparative value of microscopic detail depends upon the degree of preservation shown by the plant remains.

As previously noted, the character, composition, and forms assumed by the primary peat materials are determined by the nature of the peat-forming vegetation and vary with environmental conditions. Peat materials are readily classified as fibrous, woody, and sedimentary. The most conspicuous display of visible differences in structure and in the actual pore spaces enclosed by them is associated with fibrous types of peat, such as reed peat, sedge peat, hypnum peat, and sphagnum moss peat. Sedimentary peat materials with a high content of organic colloids derived from soft tissues of submerged and free-floating pondweeds, algae, and plankton do not show a highly developed structure; they coagulate into a sticky, plastic mass, and on air-drying contract and break into firm, compact clods. The various strengths of adhesion in all kinds of peat and muck depend largely upon the content of coagulating colloidal residues, upon the degree of decomposition (by operations of drainage or cultivation), and upon the presence of certain soluble salts (electrolytes) in ground waters.

The terms "muck" and "peat" are often used indiscriminately to designate surface organic materials and layers of peat. The term "muck" is properly used when it refers to any peat material that has been altered by drainage, aeration, the action of microorganisms, or cultivation and consequently has advanced in stage of decomposition so far that its botanical character is no

longer evident. The original parent peat material can seldom be neglected. According to its origin, muck may be designated as reed muck, sedge muck, etc., the name referring to the type of peat in an advanced degree of decomposition. Muck is a residual product that develops mainly through the activities of microorganisms; it varies in character with the nature of the peat material undergoing decomposition and with the environmental conditions under which the peat material is decomposing. Muck is usually dark brown to black, differs in the quality of its residues, is relatively low in absorbing capacity for water and soluble salts, and has a mineral content ranging between 5 and 35 per cent. In marginal portions of a peat area muck which contains more than 40 per cent of mineral matter frequently resembles the well-decomposed and mineralized organic matter in soils synthesized by microorganisms and spoken of as "humus." Peat, on the other hand, consists of relatively well recognizable plant remains; it differs in quality according to the character of the peat-forming vegetation, contains from 75 to 95 per cent of combustible organic material on a dry-weight basis, is generally brown, has a much higher water-holding capacity than its dark-colored decomposed phases, is usually poor in available nitrogen, and is deficient in potash, phosphoric acid, and other necessary mineral plant-food constituents. The term "peat" should be limited to accumulations of at least 10 inches in thickness when compact and well shrunk.¹

In regard to the general shape or main character of the primary peat materials, four principal types of structure, similar to those recommended by Nikiforoff for mineral soils (9) may be distinguished as follows: horizontal or laminated; vertical; blocky or fragmental; and granular. These similarly named aggregates of peat material, however, are not identical in appearance or origin to the aggregates of mineral soil, but such designations do serve to differentiate them.

The horizontal type of structure is characteristic of peat materials which show more or less well developed horizontal cleavage planes or lamination (plate 1, A, B, and C). The components are thin filamentous fibers, flat in shape. In most instances the aggregates are compacted and firm but easily separated from one another; upon air-drying they become brittle and crumbly. This type of structure is predominantly developed in fibrous sedge peat, reed peat, hypnum peat, and in a few varieties of sedimentary peat. Laminated or horizontal structure is particularly well defined in peat material derived from floating mats of sedges which encroach on open water in lakes and ponds. The features assumed by the peat material are due largely to the horizontal or plagiotropic growth of an interlacing network of rhizomes which push growing tips with considerable force at right angle to gravity or in an oblique position. Lateral branches and their absorbing rootlets extend rapidly in all directions, filling many cubic feet of water. This activity of rhizomes and root systems

¹ This restriction of the term complies with a ruling of the Peat Subcommittee of the International Society of Soil Science made for purposes of uniformity in surveys and mapping.

in penetrating a substratum, building up layers of peat, and likewise giving rise to structural features—aside from absorption, anchorage, and other functions—is effective not only in deep and shallow water but also in mineral soils of different texture. In spite of diverse modifying conditions in a substratum, it appears that structural features are determined to a marked extent by the inherent growth forces, activities, and character of plants and their root systems. As between the two aspects of action—between vegetation and environment—there is little doubt that the character and inherent activities of vegetation work the more profound modifications.

The vertical type of structure is characteristic of small columnar lumps and hummocklike aggregates somewhat complex and irregular in shape (plate 1, D, E, and F). The vertically elongated components can be separated with little difficulty, and under certain conditions they may assume the shape of thin flakes and plates or small fluffy aggregates. Structural aggregates of this kind are developed predominantly in fibrous sphagnum moss peat of so-called “raised moors” and in certain tussocky sedge peats of “niggerhead” bogs. Root development does not play so decisive a role as the hereditary, heliotropic upward growth of the shoots which are characteristic of these peat-forming species of plants. The root systems of cotton-grass and other sedges associated with the vegetation of raised moors are efficient binders, while the geotropically reacting, descending portions of the root system of herbaceous plants and small shrubby heaths give firm anchorage and contribute dimensional features which define the vertical structure of aggregates in deeper transitional layers of peat.

The fragmental, blocky type of structure is characteristic of woody peat materials (plate 1, G, H, and I). The aggregates are more or less complex, lumpy fragments of different sizes and amounts, irregular to angular in shape, and with a rather rough surface. Although sufficiently firm, they can be separated and, when air-dry, tend to become loose and crumbly. The structure of woody peat reflects the conditions of its development. The root systems of forest trees, shrubs, and herbaceous ground cover follow a fixed course of hereditary development and maintain a characteristic form at rather definite levels. Some species exhibit tendencies to thicken, cease absorption, and have woody flat-based roots when subjected to a high or a fluctuating water table. Woody roots do not penetrate deeply but develop rather superficially with wide reach and strongly emphasized mechanical action and buttressing effect. This entails a profound effect upon the character of the peat material (or soil) in which the trees are anchored. The accumulation of fallen timber introduces a factor of slow decomposition not present in the soft herbaceous tissue of the ground cover vegetation. As the water level drops, and as a result of shading, the consequent aeration and drainage carries with it a rapid and marked differentiation in structural aggregates, due mainly to wood-decaying fungi and synthesizing microorganisms. Their activity changes the coarser woody fragments to a granular woody residue, and the herbaceous plant tissue to

sticky colloidal constituents which bind the woody components into aggregates of varying sizes. Thus different levels in a woody layer of peat may possess their own particular structure, the aggregates of which are more or less loosely joined and vary in degree of durability and available plant nutrients. For this reason the well-drained and well-aerated woody peats derived from a dominantly deciduous swamp forest stage are the ones used, preferably, for crop production.

The granular type of structure is characteristic of sedimentary peat containing siliceous material (e.g., diatoms, wind-blown sand, volcanic ash) or calcareous material (Chara marl, shell marl); it is characteristic also of all decomposing or cultivated fibrous and woody peats generally termed "muck" (plate 1, J, K, and L). The components are more or less rounded in shape, relatively stable, and loosely connected with one another. The breaking-down process may proceed to complete pulverization. On the other hand, herbaceous sedimentary peats, derived from submerged or floating aquatic plant remains which are recognizable under the microscope and free from mineral admixtures, form a coherent, sticky plastic mass. The modifying factors are the nutrients and the quantity of water over the mineral substratum. The aggregates can be separated by saturation with water, but upon air-drying or dehydration the mass becomes hard, shrinks greatly in all dimensions, and breaks with angular fractures. The last change characterizes an irreversible condition.

With reference to the different sizes of structural aggregates, it is possible to distinguish five classes within each of the foregoing types; very coarse, coarse, medium, fine, and very fine. This range of variability within each type is, however, a function of at least two factors; relative durability and cohesion. Sizes of aggregates cannot be considered as definite, and the organic structural components cannot be characterized as yet by length of diameters of axes or thickness of aggregates as in mineral soils.

It is generally agreed in discussions concerning soil structure that the term "relative distinctness of structural aggregates" designates stability or durability and capacity to resist the destructive effects of water rather than the relative ease of separation of particular components from one another by means of tillage tools, weathering (air and temperature), pressure (compaction, shrinkage), or the influence of different salts (electrolytes). In an examination of peat and muck aggregates in the field it is possible to distinguish five grades of structural distinctness: strong, good, moderate, weak, and poor. These grades are determined mainly by the relative ability of the organic aggregates and components together to resist separation, that is, the tenacity due to the presence of coagulating organic colloids which arise generally from the decomposition of soft herbaceous tissues.

Weak structural aggregates are commonly associated with poorly decomposed coarse fibrous and woody peat materials. They are characterized by an absence of colloidal residue and, therefore, are loosely held together by a weak adhesion between the components. In sedimentary peat materials relatively

free from mineral matter, in woody and fibrous peats which have undergone decomposition, and in mixtures of sedimentary and fibrous or woody peats of intermediate texture, the grades of adhesion depend to a considerable extent upon the different percentages of colloidal matter in which the larger components are imbedded. In drained areas of peat the organic residues in solution and suspension (dopplerite, etc.) generally act as a cementing matrix, binding together granular and coarser primary peat materials. The grading of structural distinctness from strong or good to moderate and weak is regarded as strictly morphological, but it should be made on a botanical basis as well. Indeed a greater practical significance should be attached to the nature and composition of the different grades and varieties of size and shape of structural aggregates as plant remains and as physical units or elements of types of structure.

PARTICULAR CHARACTERISTICS IN THE STRUCTURE OF ALASKA PEAT MATERIAL

During the summer of 1939 the writer had an opportunity to investigate peat resources in Alaska and at that time studied the frozen peat layers of muskegs in the interior of the territory. Only one of the observations on the form of structural aggregates will be dealt with here.

Nikiforoff (8) has pointed out that freezing and melting are very influential factors in northern continental climatic regions of Europe and Asia and that they appear to have considerable effect on the development of structure.

In Alaska the thickness of frozen peat layers varies from south to north, being thicker in the north, where it reaches many feet. The depth to which the surface layers thaw during the warm but short summers depends largely upon the character of the vegetative cover and the physical properties of the peat material. During the summer, perpetually frozen peat is commonly found 20 to 50 inches below the present surface. There is every reason to believe that the origin and development of muskegs began in a relatively warm period and that freezing of these layers must be connected with climatic changes or cycles. Accordingly, the view is held that during the course of recent centuries an excess of annual cold brought about an accumulation of ice that could not be wholly melted by the limited amount of the summer's heat, and consequently frozen peat layers gradually increased in thickness with the increasing height of peat formation.

It is asserted by forest rangers that in early winter the wettest parts of the surface of a muskeg freeze first, thus getting a thicker covering of ice than the drier parts, and that in spring the ice melts first in the wetter sections of the muskeg and remains longer in the drier portions. A close study of muskegs which occur on sloping topography revealed that the periodical but irregular freezing and melting had caused unevenness and breaks in the surface, and that in some cases belts of surface peat saturated with melting water were gliding over the underlying frozen layers. This phenomenon appears to be the cause not only of many forms of water pockets but also of sharply defined transitions

in profile sections, giving rise to the development of a flaky structure in the coarser and looser peat materials. A number of slope muskegs, raised muskegs, and valley muskegs were found to have profile features consisting of several thin seams of laminated, flaky-structured aggregates. Doubtless the alternate and periodic freezing and thawing of the peat material at those levels is a result of contemporary climatic conditions. It caused the separation of platy particles and a lamination due, probably, to alternating thin lamellae of ice with lamellae of fibrous peat. Many desirable biological and chemical reactions are retarded or stopped by unfavorable low temperatures. All these affect the behavior of plants and their root systems and the character and amount of peat material accumulating as well-preserved and well-recognizable plant remains. A knowledge of the physical and chemical relations of structural aggregates is necessary for an understanding of stratigraphic features of peat land and of the interrelations existing between plants, root systems, and peat materials, and matters of proper utilization of peat deposits.

RESULTS OF PEAT INVESTIGATIONS

In summarizing briefly the results obtained from peat investigations in the last decade, it seems worth while to indicate once more that vegetation, and especially the hereditary characteristics and habits, that is, the inherent forces and functional activities, of plants and their root systems are the most important factor in determining the structural features assumed by different kinds of peat, and that the changes wrought by vegetation upon its environment, in bodies of water as well as in mineral soils, have great significance. In evaluating the suitability of an area of peat for commercial utilization or practical agriculture the term "stratification" is usually qualified by denoting the arrangement of different layers of peat below the surface, their thickness, and the character of transition from one layer to another. But stratigraphic features should be clearly distinguished from "structure" which denotes the form of individual types, kinds, and grades of primary peat materials as structural aggregates, their shape and size, degree of distinctness and tenacity, degree of decomposition and mineralization, and the properties correlated with structural aggregates which influence permeability (porosity), water-holding capacity, and other physical, chemical, and biological reactions.

The main contribution of peat investigations to soil science may, after all, be a better understanding of soil-forming processes and a better system of morphological description and classification. The essential thing is that we recognize those forces in vegetation for what they are, and appraise them for the work they are accomplishing.

REFERENCES

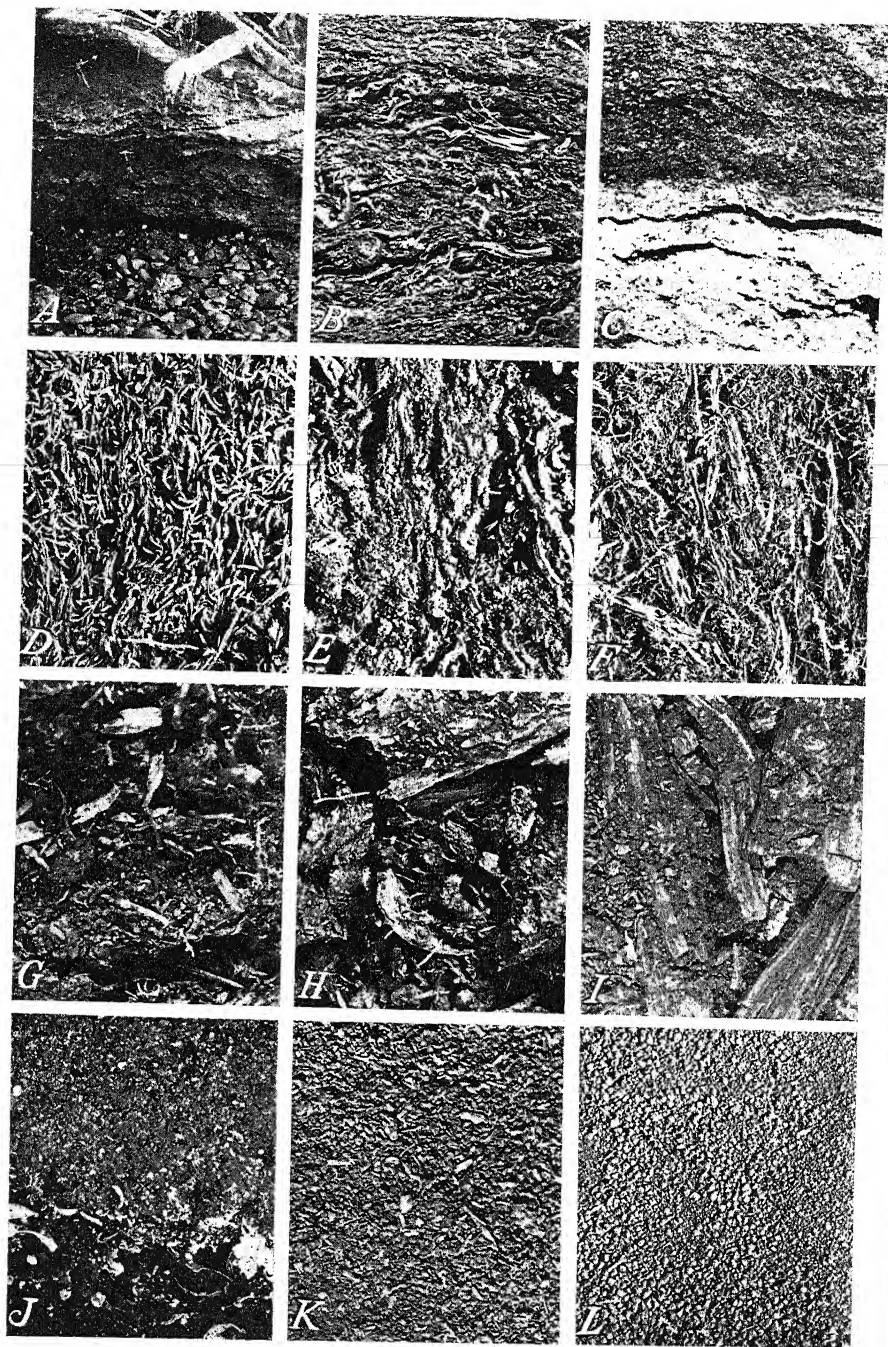
- (1) DACHNOWSKI, A. P. 1919. Quality and value of important types of peat material. U. S. Dept. Agr. Bul. 802.
- (2) DACHNOWSKI, A. P. 1926. Factors and problems in the selection of peat lands for different uses. U. S. Dept. Agr. Bul. 1419.

- (3) DACHNOWSKI-STOKES, A. P. 1929 Records of climatic cycles in peat deposits. Carnegie Institution of Washington, Report 2nd Conf. on Cycles.
- (4) DACHNOWSKI-STOKES, A. P. 1933 Grades of peat and muck for soil improvement. U. S. Dept. Agr. Cir. 290.
- (5) DACHNOWSKI-STOKES, A. P. 1933 Peat deposits in the United States. *Handbuch der Moorkunde* 7: 1-140.
- (6) DACHNOWSKI-STOKES, A. P. 1936 Peat land in the Pacific Coast States in relation to land and water resources. U. S. Dept. Agr. Misc. Pub. 248.
- (7) FEUSTEL, I. C., AND BYERS, H. G. 1930 The physical and chemical characteristics of certain American peat profiles. U. S. Dept. Agr. Tech. Bul. 214.
- (8) NIKIFOROFF, C. 1928 The perpetually frozen subsoil of Siberia. *Soil Sci.* 26: 61-81.
- (9) NIKIFOROFF, C. 1937 Method of recording soil data. *Soil Sci. Soc. Amer. Proc.* 1: 307-317.

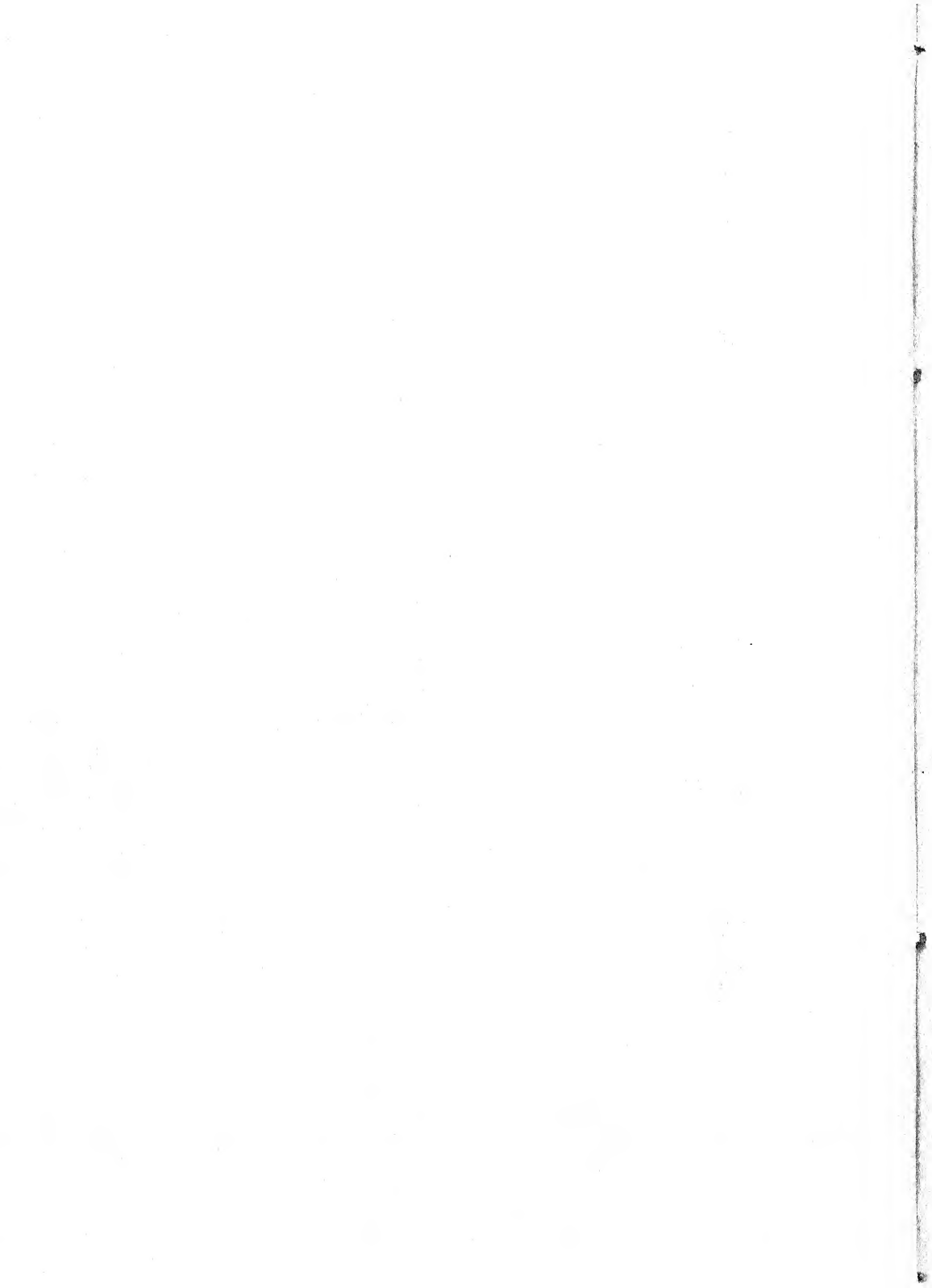
PLATE 1

PRINCIPAL TYPES OF STRUCTURAL CHARACTERISTICS OF PEAT AND MUCK

Horizontal or laminated: A, sedge peat on mineral soil; B, reed and sedge peat; C, hypnum peat on Chara marl. *Vertical:* D, sphagnum moss peat; E, tule peat; F, sedge peat. *Frag-mental or blocky:* G, ericaceous woody sedge peat; H, coniferous woody peat; I, deciduous woody peat. *Granular:* J, fine-grained sedimentary peat on shell marl; K, coarse granular and platy sedge muck; L, fine granular cultivated reed-sedge muck.



FIGS. A-L



DISSOCIATION IN AZOTOBACTER CHROOCOCCUM (BEIJERINCK)

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In connection with a study of possible symbiosis between *Azotobacter* and *Chlorella* in nitrogen fixation, we had occasion to use a culture of *A. chroococcum* from Beijerinck's original strain.¹ In plating this culture, we noticed white or cream-colored colonies occurring with black colonies on agar media. This interesting dissociation of pure culture of the organism led to a study, during the last two years, of dissociation or variation in the organism. By repeated plating we were able to isolate at one time or another at least four strains which had specific characters differing from one another. It is the object of this paper to describe these and to give some pertinent observations respecting them.

The strains isolated were as follows:

1. A black strain which was the most constant and striking. This forms colonies wholly black except as specified later. The colonies vary from 1 or 2 mm. in diameter to 20 mm. They are never more than slightly raised above the surface of the agar, are distinctly dry and nonmucilaginous, and usually, if not always, show concentric rings within the colony. Microscopically these colonies yield a picture of cells with the following description: Egg-shaped cells often in pairs, 1.2 to 1.5 μ in diameter, staining solidly and deeply with gentian violet.

2. A light brown or cinnamon-colored strain, also fairly constant which produces colonies on the average twice as large as those of strain 1. The pigment is golden brown, and the colonies show a beautiful pattern of striation from the center to the periphery of the colony, like spokes from the hub of a wheel. The colonies are seldom more than slightly raised above the surface of the agar and are as dry or nearly as dry as those of the black strain, but instead of having an entire rim, as does the black strain, are slightly more toothed or serrated. Microscopic examination yields a picture of cells of the following description: Coccuslike cells 1.0 to 1.2 μ in diameter, staining solidly and deeply with gentian violet.

3. A white strain which is fairly constant on repeated transferring but sports more readily than the black or brown strain. The colonies of this strain are large (10 or more mm. in diameter). Instead of always being circular as those of strains 1 and 2, they may spread into streamerlike masses. They

¹ Kindly furnished to us by H. Albert Barker.

are always much raised above the surface of the agar and are very wet and mucilaginous, in striking contrast to the first two strains. Occasionally a little yellowish pigment is observed. When the colonies sport, they give rise to small black colonies in the midst of the white mucilaginous mass which are identical with the black strain. Microscopically the picture obtained allows of the following description: Cells are 2 to 3.5 μ in diameter, rarely stain solidly with gentian violet but show white dots in center. Cells are mostly egg-shaped.

4. Brown mucilaginous strain. When transferred, this remains constant in respect to pigment, which is usually a cinnamon brown. In other respects, however, it has little constancy, changing quickly from very mucilaginous to fairly dry forms. The colonies are large, 10 to 15 mm. in diameter. They are irregular in contour and well raised above the surface of the agar. Microscopically the picture of this strain shows cells which are about $2\frac{1}{2}$ to 3 or 4 μ in diameter, are fairly round, but have some tendency toward the egg shape. They stain deeply and solidly with gentian violet.

There is a possibility that other strains than those described are contained within the pure culture known as *A. chroococcum*. For example, one strain, which we formerly called "pearly white colony," seems to be distinct from the foregoing, but on constant transferring, it develops a very light brown pigment and loses some of its original dryness. We have, therefore, not decided as yet to call it an independent additional strain to those described. One of the striking characteristics of *A. chroococcum* in the parent culture under study here, as well as in the strains separated therefrom, is the constant tendency to sporting or variation, which characterizes them. Even strain 1 may, after several platings in which dry black colonies appear exclusively, suddenly develop a small gelatinous nipple usually at the edge of one of the colonies, from which may again be segregated gelatinous and dry black forms. The same is true of the white mucilaginous forms and of the others. The appearance of sports or variants of strain 3 is indicated by the appearance of black dots on the white mucilaginous masses. These dots will give rise to the dry black form, strain 1. It is also notable that some of the strains apparently contain some lethal factor which kills them after one or two transfers. For example, this is true of the white mucilaginous form, which in many instances could not be made to grow after the first or second transfer.

PHYSIOLOGICAL ASPECTS OF THE VARIANTS OF *AZOTOBACTER CHROOCOCCUM* BEIJERINCK STRAIN

Our studies on the physiological aspects of this problem have been very limited. The nitrogen-fixing power of the various strains was studied, however, on a considerable scale. In these studies two long series of cultures were grown. In one, the carbohydrate source consisted of a mixture of mannite, dextrose, and sucrose in a concentration of 1 per cent of the solution. In the other, carbon was furnished in the form of ethyl alcohol in a concentration of

1 per cent. The separate cultures within a given strain were found to be highly variable in their nitrogen-fixing power. This variability may be higher than 100 per cent within the strain. In general, all the strains fix more nitrogen on alcohol than they do on the mixed carbohydrate used as the alternative medium. Nevertheless, it is true that the original culture of *A. chroococcum* is from 50 to 100 per cent more efficient as a nitrogen fixer than any of the strains derived from it. The absolute amounts of nitrogen fixed varied from 2 to $2\frac{1}{2}$ mgm. per gram of carbon source in the first medium and from 3 to $4\frac{1}{2}$ mgm. in the second medium. The fixation of nitrogen for the original culture varied from $4\frac{1}{2}$ to $5\frac{1}{2}$ mgm. of nitrogen per gram of carbon source.

The only other aspect of our physiological observations upon which we may properly comment here is that of pigment forms. We have never seen in *Azotobacter* a pigment so black as that in strain 1. The pigment is not glistening but is dry and coallike. The question naturally arises: What is the mechanism within the cells of a given strain which causes a certain chemical reaction in its metabolism to veer in the direction of making this particular pigment, about the chemical nature of which we know relatively little? The further question is: Why do some cells segregate from a pure culture in a form which apparently is entirely lacking in the mechanism or the catalyst for black or even brown pigment formation? It should be recalled in connection with these discussions that many years ago Sackett proved that *A. chroococcum*, which normally produces black pigment to a limited extent, can be made to produce it to a large degree when supplied with nitrate in the medium. It will also be recalled that Winogradsky only a few years ago showed that the presence of benzoic acid or of benzoates in the medium is conducive to the formation of black pigment by *A. chroococcum*. The reader will note, however, that in our experiments we used no nitrate or benzoate in the medium. Our medium consisted of the usual salt mixture employed in Ashby's medium with the carbon source as explained above. It appears, therefore, that the device used by Sackett or the one used by Winogradsky is more in the nature of a stimulus to the greater development of the black strain than of a stimulus to the formation of pigment by all the cells of a culture of *A. chroococcum*. It will be observed again that we have isolated a black strain which is more intense in its pigment formation than any of which we have knowledge, and this is accomplished without the use of any stimulating substance whatever.

PLATE 1

FIG. 1. Original culture of *A. chroococcum* from Beijerinck strain showing ordinary segregation of colonies. $\frac{1}{2}$ Natural size.

FIG. 2. Colonies of the black strain 1. $\frac{1}{2}$ Natural size.

FIG. 3. Cell form of the black strain. $\times 480$.

FIG. 4. Colonies of the dry brown strain 2. $\times 2$.

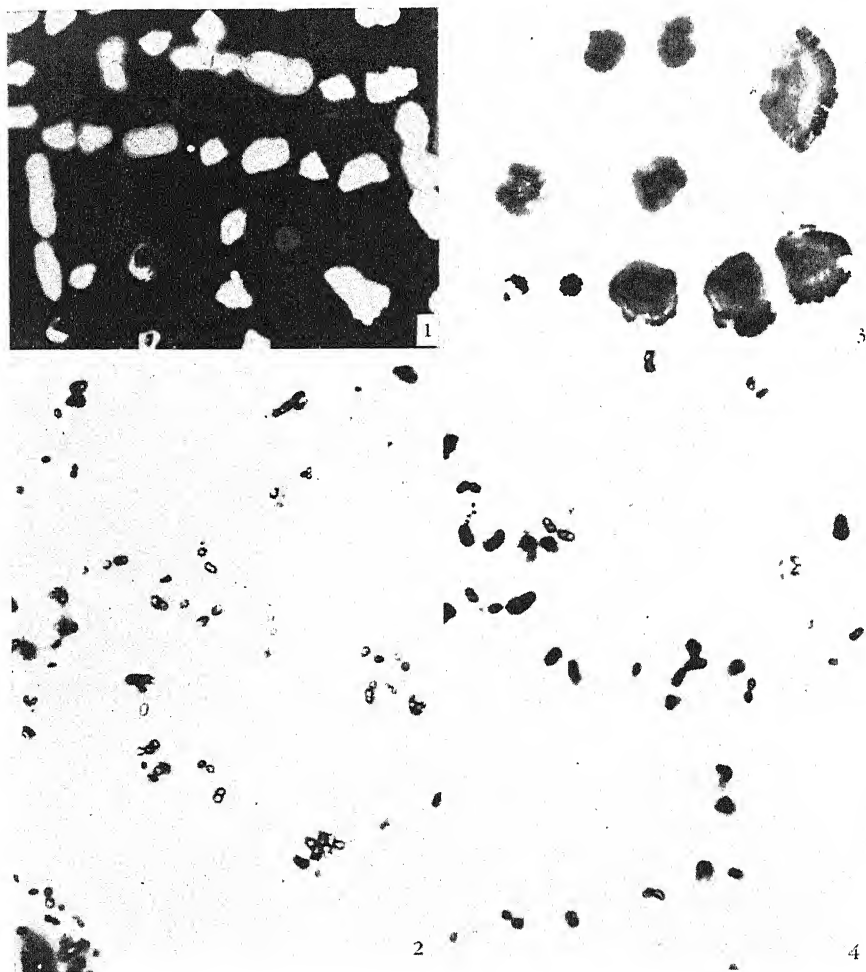
FIG. 5. Cell form of the dry brown strain. $\times 480$.



FIGS. 1-5

PLATE 2

- FIG. 1. Colonies of the white mucilaginous strain 3. $\times 2$.
FIG. 2. Cell form of the white mucilaginous strain. $\times 480$.
FIG. 3. Colonies of the brown mucilaginous strain 4. $\times 2$.
FIG. 4. Cell form of the brown mucilaginous strain. $\times 480$.



FIGS. 1-4

THE NUTRITIONAL REQUIREMENTS OF SOIL BACTERIA—A BASIS FOR DETERMINING THE BACTERIAL EQUILIBRIUM OF SOILS¹

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In reviewing recent progress in the field of bacterial nutrition, one is impressed by the variation in the requirements of the numerous organisms which have been studied. This is illustrated in table 1, indicating the diversity in growth factor requirements established for various species of bacteria. It is this fact, together with an appreciation of the great amount of labor involved in defining the needs of even a single organism, that makes the subject of nutritional requirements of soil bacteria, as a whole, seem highly complex. In studying the physiology of soil isolates as described herein, however, it soon became apparent that, in spite of the variability in biochemical behavior noted even with many types closely related morphologically (43), remarkable similarities in nutritive requirements exist among the many members of the soil population. Moreover, it has been found possible to meet the growth requirements of the majority of the cultivable soil organisms with purely synthetic media. Utilization of fundamental nutritional differences as a basis for grouping these organisms has provided a useful method for following changes in the bacterial equilibrium of soils.

The generalities expressed in this paper are based upon individual nutritional studies of over three thousand isolates obtained from various soils in connection with work on root-rot disease of strawberry. Since space does not permit a consideration of each organism separately, the general plan of approach is presented, together with a few selected examples showing the application of nutritional data to soil balance investigations.

EXPERIMENTAL

Nutritional requirements of soil bacteria

All bacteria used in nutritional studies were obtained by plating on a non-selective soil extract medium according to previously described methods (18, 43). Colonies were systematically picked so that all on a whole or definite sector of a plate, regardless of size or relative abundance, were included in the

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sample taken for qualitative analysis. These organisms were inoculated into tubes of basal medium (see below) containing 1.0 gm. powdered Difco yeast extract per liter (medium Y), and allowed to develop for 5 days at 28°C. Morphology and Gram-stain reaction of the bacteria were determined by microscopic observation of smears from these cultures.

TABLE 1
Review of recent work in bacterial nutrition

SPECIES	GROWTH FACTORS	REFERENCES
<i>Staphylococcus aureus</i>	Nicotinic acid, thiamin, uracil, biotin	(9, 10, 12, 15, 31, 33)
<i>Staphylococcus albus</i>	Nicotinic acid, thiamin, vitamin B ₆	(40)
<i>Sarcina flava</i>	Thiamin, riboflavin, ascorbic acid	(34)
<i>Streptococcus hemolyticus</i>	Pantothenic acid, riboflavin, vitamin B ₆ , glutamine	(7, 17, 20, 21, 52, 53)
<i>Escherichia coli</i>	Indole acetic acid	(1, 2)
<i>Proteus vulgaris</i>	Nicotinic acid	(6)
<i>Proteus morganii</i>	Pantothenic acid, nicotinic acid	(30)
<i>Shigella dysenteriae</i>	Nicotinic acid	(3, 16)
<i>Brucella abortus</i>	Nicotinic acid, thiamin	(8)
<i>Photobacterium phosphorescens</i>	Riboflavin	(4)
<i>Hemophilis influenzae</i>	Hematin, co-enzyme	(19)
<i>Pasteurella pestis</i>	(Proline, phenylalanine, cystine)	(32)
<i>Rhizobium trifolii</i>	Biotin, thiamin	(27, 28, 48, 49, 50)
<i>Corynebacterium diphtheriae</i>	β -alanine, nicotinic acid, pimelic acid	(23, 24, 25, 26)
<i>Lactobacillus delbrueckii casei</i>	Pantothenic acid, nicotinic acid, riboflavin, vitamin B ₆	(22, 36, 37, 38)
<i>Propionobacterium pentosaceum</i>	Thiamin, pantothenic acid, nicotinic acid	(42, 51)
<i>Clostridium acetobutylicum</i>	Biotin, (asparagine)	(39, 41, 44, 45)
<i>Clostridium sporogenes, botulinum</i>	"Sporogenes vitamin"	(5, 11, 29)

On the basis of preliminary tests with a limited number of isolates (47), three synthetic media, as follows, were developed for use in this work:

Basal medium consisting of glucose, 1.0 gm.; K₂HPO₄, 1.0 gm.; KNO₃, 0.5 gm.; MgSO₄, 0.2 gm.; CaCl₂, 0.1 gm.; NaCl, 0.1 gm.; FeCl₃, 0.01 gm.; distilled water, 1 liter. Heat to 100°C., filter and adjust reaction to pH 6.8. *Medium B.*

Amino acid medium consisting of basal medium plus cysteine, 0.05 gm.; alanine, 0.1 gm.; proline, 0.05 gm.; asparagine, 0.05 gm. per liter. *Medium A.*

Growth factor medium consisting of basal medium plus cysteine, 0.05 gm.; thiamin, 100 gammas; biotin concentrate (purity 0.4 per cent), 0.02 cc. per liter. *Medium G.* Biotin concentrate was fractionated from egg yolk by the procedure of K \ddot{u} gl and Tonnies (14) and standardized by three separate assays (13, 35, 50).

Each isolate was transferred by loop inoculum from medium Y into media B, A, and G and incubated 5 days at 28°C. The growth response of each organism in the three synthetic media was recorded as follows, each isolate being considered separately: The tube showing heaviest growth was assigned a turbidity value of 4, and other cultures of that organism were rated by comparison. If multiplication occurred only in the growth factor medium, the reaction of the organism in the three media B, A, and G was abbreviated 0-0-4. If equally heavy growth resulted in all three media, the reaction was expressed 4-4-4. In a case where heaviest development occurred in medium A, but only about one half the growth was observed in medium G and about one quarter the amount in medium B, the notation would be 1-4-2, etc. No effort was made to distinguish between slight differences. Only readily discernible variations in growth response were recorded as significant.

On the basis of their reactions in the three synthetic media, all of the soil organisms studied fell into one of the following five major divisions:

A. Bacteria with simple requirements. These bacteria are capable of synthesizing all substances necessary for their maximum growth from the simple ingredients of the glucose-nitrate-salts medium. Their growth could not be significantly improved by the substitution of more complex media.

B. Bacteria which require cysteine. For their maximum development, these bacteria require the addition of cysteine to the basal medium and therefore grow equally well in media A and G. Some isolates are capable of limited growth in medium B, whereas others produce no visible turbidity at all in the absence of cysteine. Thioglycollic acid exerts a stimulating effect on some members of this group, but in no case has it been observed to replace completely the amino acid.

C. Bacteria which require a mixture of amino acids. As reported elsewhere (47), no single amino acid is capable of fully stimulating this group of bacteria. The important member of the essential combination is cysteine, which to be stimulative at all in many cases, must be present together with alanine, proline (hydroxyproline less effective), or asparagine. All organisms of this group that have been studied show the same specificity. The only differences noted relate to the activity of cysteine alone: some organisms do not respond at all to the single amino acid, whereas others do show a submaximal response. Of those which exhibit some growth in the presence of cysteine alone, a few also have the power of growing slowly in the basal medium. The reason why alanine, proline, or asparagine is highly effective while other closely related substances are virtually inert has not been determined. Although cysteine plus any one of the other three substances would probably be satisfactory for most organisms of this class, all three compounds are included in medium A.

D. Bacteria which require growth factors. These bacteria produce their maximum growth in medium G. It has been pointed out (47) that organisms of this class are highly specific in their growth factor requirements. They

respond either to thiamin or to biotin or to both, but to no other growth substances that have been tested. Moreover, the response occurs only in the presence of cysteine. As is the case in the amino acid group, certain of these organisms will grow at a submaximal rate when supplied with cysteine alone. Of these there are some which develop to a limited extent even on medium B, but for all members of this group, one or other or both of the growth substances is necessary for full development.

E. Bacteria with undefined requirements. Although the majority of soil isolates can be grouped on the basis of their nutrition into one of the foregoing classes, there is invariably a smaller number which have more complex requirements and therefore fail to grow in medium B, A, or G. This group has not been closely studied but appears to be a heterogeneous one. Some of the more fastidious organisms grow only on the soil extract plates and fail to develop on subsequent transfer to medium Y. Others grow well in medium Y but not in any of the differential synthetic media.

Classification of soil bacteria on the basis of nutritional requirements

In order to interpret the large number of data obtained in nutritional studies of representative bacteria from any particular soil, some scheme for condensation of results is essential. The following system for grouping the organisms on the basis of their nutritional requirements was planned to provide a simple, workable method by which the qualitative data could be conveniently summarized. The figures in brackets represent growth responses of the organisms in media B, A, and G. Any organism isolated from the soil can be placed in one and only one of ten recognized groups. From such grouping, the percentage occurrence of the various nutritional types may be readily calculated.

A. Bacteria capable of synthesizing all substances for their maximum growth from constituents of a glucose-nitrate-salts medium.

Gram-positive and Gram-variable bacteria.....	Group 1	{	[444, 434, 433, 334, 343, 344, 414, etc.]
Gram-negative "fluorescens" type (producing pyocyanin).....	Group 2		
Other Gram-negative bacteria.....	Group 3		

B. Bacteria requiring the addition of cysteine to basal medium for maximum growth.

Capable of submaximal growth in medium B.....	Group 4	[144, 244]
Unable to grow in medium B.....	Group 5	[044]

C. Bacteria requiring the addition of cysteine plus alanine, proline, or asparagine for maximum growth.

Capable of submaximal growth in medium B.....	Group 6	[140, 141, 142, 143, 240, 241, 242, 243]
Unable to grow in medium B.....	Group 7	[040, 041, 042, 043]

D. Bacteria requiring the addition of cysteine plus thiamin and/or biotin for maximum growth.

Capable of submaximal growth in medium B.....Group 8 [114, 124, 134, 214, 224, 234]

Unable to grow in medium B.....Group 9 [004, 014, 024, 034]

E. Bacteria incapable of growth in any above synthetic media, requiring the presence of unidentified substances in soil extract or yeast extract.

Group 10 [000]

The bacterial equilibrium of soils

The availability of growth essentials is obviously a fundamental factor in determining the interrelationships among the many members of the complex soil population. In every soil investigated a balance exists between two general nutritional classes of bacteria. On the one hand, are bacteria capable of rapid growth on a simple substrate, and in opposition to them are others dependent for their development on an ample supply of certain specific compounds. If conditions favor an increase in the occurrence of one group, the incidence of the other must correspondingly fall. This microbiological equilibrium may be conveniently expressed on a numerical basis, if the percentage occurrence of group 3 bacteria is assigned a negative value, and the incidence of groups 5, 7, and 9 are given positive values. The sum of these is designated the "bacterial balance index." Those specific groups selected for this calculation possessed, in addition to nutritive differences, a further significance in that group 3 bacteria showed a consistently higher incidence in the root-rot soils and groups 5, 7, and 9 in the healthy soils examined. On the other hand, the incidence of groups 1, 2, 4, 6, 8, and 10 did not appear to be related to the condition of the soil.

There are many factors which may cause the bacterial balance index to rise or fall. It is beyond the scope of this paper to deal with them as related to any specific soil problem. As examples, however, of the application of the method to soil balance studies, data are presented to show the changes which can be observed to result from the influence of (a) seasonal factors, and (b) plant growth.

Example 1. Influence of seasonal factors on the bacterial equilibrium of a soil. Soil samples were taken at approximately monthly intervals from between the rows in a strawberry plantation. The soil was the common light sandy loam of the district which had received dressings of barnyard manure. One square foot of soil to a depth of about 6 inches was thoroughly mixed and 10 gm. removed for bacteriological analysis. There was no plant growth on the area selected.

The nutritional requirements of a representative number of organisms from April, May, and July isolations are shown in table 2. The analysis of these data and the calculation of the bacterial balance index for each sample are

TABLE 2

Nutritional requirements of representative bacteria from field soil at different times of the year

CULTURE NUMBER	APRIL 27			MAY 27			JULY 6		
	Medium B	Medium A	Medium G	Medium B	Medium A	Medium G	Medium B	Medium A	Medium G
1	2	4	3	0	0	0	0	0	4
2	0	3	4	0	1	4	Actinomycete		
3	4	4	4	0	4	2	0	3	4
4	0	0	0	0	0	0	0	0	4
5	0	0	4	Actinomycete			0	3	4
6	3	4	4	0	0	0	0	0	4
7	0	0	0	0	0	0	1	2	4
8	No growth in Med. Y			0	2	4	0	1	4
9	0	1	4	Actinomycete			3	3	4
10	0	1	4	No growth in Med. Y			4	4	4
11	1	2	4	0	4	4	0	4	2
12	0	4	0	Actinomycete			Actinomycete		
13	0	1	4	0	3	4	Actinomycete		
14	0	4	0	0	3	4	2	2	4
15	3	4	4	0	4	0	3	3	4
16	0	4	0	0	3	4	3	4	3
17	0	0	0	0	1	4	0	0	0
18	0	0	0	0	4	4	3	3	4
19	0	2	4	0	0	0	0	0	4
20	2	1	4	No growth in Med. Y			4	2	3
21	0	2	4	0	4	2	4	2	3
22	0	4	2	No growth in Med. Y			0	1	4
23	0	0	0	0	4	4	Actinomycete		
24	2	2	4	0	4	2	0	2	4
25	3	4	3	0	4	4	1	4	2
26	Actinomycete			0	4	4	0	4	4
27	0	1	4	No growth in Med. Y			3	2	4
28	4	4	4	4	4	4	3	4	4
29	0	0	0	4	4	4	4	4	4
30	2	4	4	No growth in Med. Y			2	4	4
31	0	0	4	4	3	4	3	4	4
32	0	0	0	4	1	4	3	3	4
33	0	0	0	0	3	4	0	1	4
34	No growth in Med. Y			3	4	4	4	3	3
35	1	3	4	3	4	4	4	2	0
36	0	3	4	0	2	4	No growth in Med. Y		
37	0	4	2	0	3	4	4	4	4
38	0	0	0	0	0	0	0	0	0
39	No growth in Med. Y			No growth in Med. Y			3	4	4
40	4	4	4	1	1	4	3	4	4
41	0	2	4	No growth in Med. Y					
42	0	1	4						
43	4	4	4						

shown in table 3. It will be observed that little change occurred in the index for the first two months, but by July it had fallen considerably. The data indicate that those bacteria capable of growth on the simpler substrates found conditions increasingly more favorable to their development while the reverse was true for the more fastidious types. It is apparent, therefore, that the bacterial equilibrium of an ordinary soil does not remain constant throughout

TABLE 3
Seasonal shift in the bacterial equilibrium of a field soil

NUTRITIONAL GROUP	APRIL 27			MAY 27			JULY 6		
	Culture number	Total number	Per cent	Culture number	Total number	Per cent	Culture number	Total number	Per cent
1	3, 28	2	4.8	28, 29	2	5.2	15, 16, 21, 27, 29, 34, 35, 37	8	21.6
2	15	1	2.4		0	0		0	0
3	6, 25, 40, 43	4	9.6	3, 32, 35, 34	4	10.4	9, 10, 18, 20, 28, 31, 32, 39, 40	9	24.3
4	30	1	2.4		0	0	30	1	2.7
5		0	0	11, 18, 29, 25, 26	5	13.0	26	1	2.7
6	1	1	2.4		0	0	25	1	2.7
7	12, 14, 16, 27, 37	5	12.0	3, 15, 21, 24	4	10.4	11	1	2.7
8	11, 20, 24, 35	4	9.6	40	1	2.6	7, 14	2	5.4
9	2, 5, 9, 10, 13, 19, 21, 27, 31, 36, 41, 42	12	28.8	2, 8, 13, 14, 16, 17, 33, 36, 37	9	23.4	1, 3, 4, 5, 6, 8, 19, 22, 24, 33	10	27.0
10	4, 7, 8, 17, 18, 23, 29, 32, 33, 34, 38, 39	12	28.8	1, 4, 6, 7, 10, 19, 20, 22, 27, 30, 38, 39, 41	13	33.8	17, 36, 38	3	8.1
	Bacterial balance index = -10+0+12+29 = +31			Bacterial balance index = -10+13+10+23 = +36			Bacterial balance index = -24+3+3+27 = +9		

the growing period but undergoes rather striking modification as the season progresses.

Example 2. Influence of plant growth on the bacterial equilibrium of a soil. The bacterial flora at the root surfaces of certain growing plants differs qualitatively as well as quantitatively from that away from the influence of the root (18, 47). As previously described, those organisms requiring growth factors are selectively stimulated at the root surface, an effect which is explained, at

TABLE 4

Nutritional requirements of bacteria from rhizosphere soil and from soil beyond the influence of the root

CULTURE NUMBER	RHIZOSPHERE SOIL			CONTROL SOIL		
	Medium B	Medium A	Medium G	Medium B	Medium A	Medium G
1	0	0	0	0	0	0
2	0	0	4	0	4	1
3	1	2	4	4	4	4
4	0	0	4	0	0	0
5	3	3	4	2	4	4
6	0	1	4	Actinomycete		
7	0	4	0	0	0	0
8	0	0	0	1	1	4
9	0	4	0	0	3	4
10	0	0	4	0	1	4
11	0	4	4	0	0	4
12	0	1	4	0	0	4
13	0	2	4	0	2	4
14	4	4	4	0	0	4
15	No growth in Medium Y			0	2	4
16	0	0	0	0	4	1
17	0	4	0	0	4	4
18	0	1	4	0	0	4
19	3	4	3	3	4	4
20	0	2	4	4	4	4
21	0	0	4	0	0	0
22	0	1	4	0	0	0
23	0	1	4	3	3	4
24	0	1	4	3	4	4
25	0	4	0	3	4	4
26	0	4	4	0	0	4
27	0	0	4	0	0	4
28	0	2	4	4	3	4
29	0	3	4	0	0	4
30	0	2	4	0	0	4
31	0	2	4	0	0	0
32	3	3	4	0	0	0
33	0	1	4	4	4	4
34	0	0	0	No growth in Medium Y		
35	0	0	4	0	0	0
36	0	4	0	Actinomycete		
37	0	4	3	3	4	3
38	0	0	4	0	0	4
39	0	0	4	4	4	3
40	4	2	3	4	3	3
41	0	0	0	3	4	4
42	0	2	4	4	4	4
43	0	2	4	0	4	4
44	0	1	4	0	0	0
45	0	4	4			
46	0	4	0			
47	1	4	3			

least in part, by the liberation of small but significant quantities of the necessary factors by the roots (46). It appears that this "rhizosphere effect" is virtually confined to the actual root surface, any soil not in contact with the roots themselves being unaffected. Even though this modified zone is limited in extent, however, it is nevertheless of importance to the plant, for it is in this environment that the roots perform their functions. Data illustrating the characteristic shift in the bacteriological equilibrium which normally occurs at root surfaces of healthy Premier strawberry seedlings are presented in tables 4 and 5. The bacterial balance index of the rhizosphere soil is markedly higher than that of the control soil, indicating that those organisms possessing the

TABLE 5
Modification of the bacterial equilibrium in the vicinity of plant roots

NUTRI- TIONAL GROUP	RHIZOSPHERE			CONTROL		
	Culture number	Total number	Per cent	Culture number	Total number	Per cent
1	19	1	2.1	3, 28, 33, 39, 40, 41, 42	7	16.1
2		0	0	19, 20, 23, 25	4	9.2
3	5, 14, 32, 40	4	8.6	24, 37	2	4.6
4		0	0	5	1	2.3
5	11, 26, 45	3	6.5	17, 43	2	4.6
6	47	1	2.1		0	0
7	7, 9, 17, 25, 36, 37, 46	7	15.1	2, 16	2	4.6
8	3	1	2.1	8	1	2.3
9	2, 4, 6, 10, 12, 13, 18, 20, 21, 22, 23, 24, 27, 28, 29, 30, 31, 33, 35, 38, 39, 42, 43, 44	24	50.4	9, 10, 11, 12, 13, 14, 15, 18, 26, 27, 29, 30, 38	13	29.9
10	1, 8, 15, 16, 34, 41	6	13.0	1, 4, 7, 21, 22, 31, 32, 34, 35, 44	10	23.0
	Bacterial balance index = -9+7+15+50 = +63			Bacterial balance index = -5+5+5+30 = +35		

more complex nutritive requirements are favored in the immediate vicinity of the roots.

DISCUSSION

Soil investigations have been hampered by the lack of satisfactory qualitative methods for studying the bacterial flora. Quantitative data alone, though indicating numerical trends, are inadequate when information regarding the nature of the organisms is desired. Nor do isolated physiological tests of "nitrate-reducing power," "gelatin liquefaction," "action on milk," etc., materially clarify the picture. The fact that the balance between various members of the soil population at any given time depends on the availability of compounds essential for the growth of those organisms is self-evident. It is

believed, therefore, that a qualitative approach to the study of soil bacteria based on a knowledge of the substances controlling the multiplication of those organisms is fundamentally sound. The method suggested herein has proved to be a workable one capable of yielding interesting results when applied to specific soil problems. Very probably the details of procedure may be considerably improved upon or modified to suit various purposes, but it is felt that the general principles concerned will provide a useful tool for qualitative investigations of the complex bacterial flora of the soil.

SUMMARY

It has been found possible to cultivate on various synthetic media the majority of soil bacteria capable of being isolated by nonselective plating methods. By classifying the organisms according to their nutritional needs, as determined by their growth response in media of varying complexity, a useful approach to the qualitative study of soil populations has been developed.

In any given soil, an equilibrium exists between bacteria with simple growth requirements and bacteria with more complex ones. This balance may be modified by many factors. The effects of seasonal influences and plant growth are given as examples to show how, by application of the method, changes in the "bacterial balance index" of soils may be observed.

REFERENCES

- (1) BALL, E. 1938 Hetero-auxin and the growth of *E. coli*. *Jour. Bact.* 36: 559-565.
- (2) BECKWITH, T. D., AND GEARY, E. M. 1940 The effect of indole-3-acetic acid on multiplication of *E. coli* and *E. typhosa*. *Jour. Infect. Diseases* 66: 78-79.
- (3) DORFMAN, A., KOSER, S. A., AND SAUNDERS, F. 1938 The activity of certain nicotinic acid derivatives as growth essentials for the dysentery bacillus. *Jour. Amer. Chem. Soc.* 60: 2004.
- (4) DOUDOROFF, M. 1938 Lactoflavin and bacterial luminescence. *Enzymologia* 5: 239-243.
- (5) FILDES, P. 1935 The tryptophan and "sporogenes vitamin" requirements of *B. botulinus*. *Brit. Jour. Exp. Path.* 16: 309-314.
- (6) FILDES, P. 1938 The growth of *Proteus* on ammonium lactate plus nicotinic acid. *Brit. Jour. Exp. Path.* 19: 239-244.
- (7) FILDES, P., AND GLADSTONE, G. P. 1939 Glutamine and the growth of bacteria. *Brit. Jour. Exp. Path.* 20: 334-341.
- (8) KERBY, G. P. 1939 Nicotinic acid and thiamin hydrochloride as growth promoting factors for *Brucella*. *Jour. Bact.* 37: 495-499.
- (9) KNIGHT, B. C. J. G. 1937 The nutrition of *Staphylococcus aureus*; nicotinic acid and vitamin B₁. *Biochem. Jour.* 31: 731-737.
- (10) KNIGHT, B. C. J. G. 1937 The nutrition of *Staphylococcus aureus*. The activities of nicotinamide, aneurin, and related compounds. *Biochem. Jour.* 31: 966-973.
- (11) KNIGHT, B. C. J. G., AND FILDES, P. 1933 A vitamin necessary for the growth of *B. sporogenes*: its relation to auxin and other growth factors. *Brit. Jour. Exp. Path.* 14: 112-124.
- (12) KNIGHT, B. C. J. G., AND McILWAIN, H. 1938 The specificity of aneurin and nicotinamide in the growth of *Staphylococcus aureus*. *Biochem. Jour.* 32: 1241-1251.

- (13) KÖGL, F., AND FRIES, N. 1937 Über den Einfluss von Biotin, Aneurin und Meso-inosit auf das Wachstum verschiedener Pilzarten. *Ztschr. Physiol. Chem.* 249: 93-110.
- (14) KÖGL, F., AND TONNIS, B. 1936 Über das Bios-Problem. Darstellung von kristallisiertem Biotin aus Eigelb. *Ztschr. Physiol. Chem.* 242: 43-73.
- (15) KÖGL, F., AND WAGTENDONK, W. S. VAN 1938 Significance of biotin in the growth of *Staphylococcus pyogenes aureus*. *Rec. Trav. Chim.* 57: 747-754.
- (16) KOSER, S. A., DORFMAN, A., AND SAUNDERS, F. 1938 Nicotinic acid as an essential growth substance for dysentery bacilli. *Proc. Soc. Exp. Biol. and Med.* 38: 311-313.
- (17) LANDY, M. 1939 Influence of glutamine on the growth of *Streptococcus haemolyticus*. *Nature* 144: 512-513.
- (18) LOCHHEAD, A. G. 1940 Qualitative studies of soil micro-organisms: III. Influence of plant growth on the character of the bacterial flora. *Canad. Jour. Res. (C)* 18: 42-53.
- (19) LWOFF, A., AND LWOFF, M. 1937 Rôle physiologique de l'hémimine pour *H. influenzae* Pfeiffer. *Ann. Inst. Pasteur* 59: 129-136.
- (20) McILWAIN, H. 1939 The specificity of glutamine for the growth of *Streptococcus haemolyticus*. *Biochem. Jour.* 33: 1942-1946.
- (21) McILWAIN, H., et al. 1939 Glutamine and the growth of *Streptococcus hemolyticus*. *Biochem. Jour.* 33: 223-229.
- (22) MUELLER, E. F. 1938 Vitamin B₆ (adermin) as a growth factor for lactic acid bacteria. *Ztschr. Physiol. Chem.* 254: 285-286.
- (23) MUELLER, J. H. 1937 Studies on cultural requirements of bacteria. Pimelic acid as a growth stimulant for *C. diphtheriae*. *Jour. Bact.* 33: 163-178.
- (24) MUELLER, J. H. 1937 Nicotinic acid as a growth accessory substance for the diphtheria bacillus. *Jour. Bact.* 34: 429-441.
- (25) MUELLER, J. H., AND COHEN, S. 1937 β -alanine as a growth accessory for the diphtheria bacillus. *Jour. Bact.* 34: 381-386.
- (26) MUELLER, J. H., AND KLOTZ, A. W. 1938 Pantothenic acid as a growth factor for the diphtheria bacillus. *Jour. Amer. Chem. Soc.* 60: 3086.
- (27) NILSSON, R., BJALFVE, G., AND BURSTROM, D. 1938 Vitamin B₁ als Wachsfaktor für *Bact. radicolola*. *Naturwissenschaften* 18: 284.
- (28) NILSSON, R., BJALFVE, G., AND BURSTROM, D. 1938 Growth factors for *Bact. radicola*: II. *Lantbr. Högskolans. Ann. [Sweden]* 6: 299-326.
- (29) PAPPENHEIMER, A. M. 1935 The nature of the "sporogenes vitamin," an essential growth factor for *Cl. sporogenes* and related organisms. *Biochem. Jour.* 29: 2057-2063.
- (30) PELCZAR, M. J., JR., AND PORTER, J. R. 1940 Pantothenic acid and nicotinic acid as essential growth substances for Morgan's bacillus, *Proteus morganii*. *Proc. Soc. Exp. Biol. and Med.* 43: 151-154.
- (31) PORTER, J. R., AND PELCZAR, M. J. JR. 1940 Biotin (bios IIB, vitamin H)—an essential growth factor for certain staphylococci. *Science* 91: 576-577.
- (32) RAO, M. S. 1939 The nutritional requirements of the plague bacillus. *Indian Jour. Med. Res.* 27: 75-89.
- (33) RICHARDSON, G. M. 1936 The nutrition of *Staphylococcus aureus*. Necessity for uracil in anaerobic growth. *Biochem. Jour.* 30: 2184-2190.
- (34) SARTORY, A., SARTORY, R., AND MEYER, J. 1938 The influence of certain water-soluble vitamins on the velocity of growth of various cocciform bacteria. *Compt. Rend. Acad. Sci. [Paris]* 206: 1414.
- (35) SNELL, E. E., EAKIN, R. E., AND WILLIAMS, R. J. 1940 A quantitative test for biotin and observations regarding its occurrence and properties. *Jour. Amer. Chem. Soc.* 62: 175-178.

- (36) SNELL, E. E., AND PETERSON, W. H. 1940 Growth factors for bacteria: X. Additional factors required by certain lactic acid bacteria. *Jour. Bact.* 39: 273-285.
- (37) SNELL, E. E., STRONG, F. M., AND PETERSON, W. H. 1938 Pantothenic and nicotinic acids as growth factors for lactic acid bacteria. *Jour. Amer. Chem. Soc.* 60: 2825.
- (38) SNELL, E. E., STRONG, F. M., AND PETERSON, W. H. 1939 Growth factors for bacteria: VIII. Pantothenic and nicotinic acids as essential growth factors for lactic and propionic acid bacteria. *Jour. Bact.* 38: 293-308.
- (39) SNELL, E. E., AND WILLIAMS, R. J. 1939 Biotin as a growth factor for the butyl alcohol producing anaerobes. *Jour. Amer. Chem. Soc.* 61: 3594.
- (40) SPIES, T. D., AND VILTER, G. P. 1940 Vitamin B₆ as an accessory growth factor for *Staphylococcus albus*. *Science* 91: 200-201.
- (41) TATUM, E. L., PETERSON, W. H., AND FRED, E. B. 1934 An unknown factor stimulating the formation of butyl alcohol by certain butyric acid bacteria. *Jour. Bact.* 27: 207-217.
- (42) TATUM, E. L., WOOD, H. G., AND PETERSON, W. H. 1936 Growth factors for bacteria: V. Vitamin B₁, a growth stimulant for propionic acid bacteria. *Biochem. Jour.* 30: 1898-1904.
- (43) TAYLOR, C. B., AND LOCHHEAD, A. G. 1938 Qualitative studies of soil micro-organisms: II. A survey of the bacterial flora of soils differing in fertility. *Canad. Jour. Res. (C)* 16: 162-173.
- (44) WEIZMANN, C. H., AND ROSENFELD, B. 1937 Activation of the butanol-acetone fermentation of carbohydrates by *Clostridium acetobutylicum*. *Biochem. Jour.* 31: 619-639.
- (45) WEIZMANN, C. H., AND ROSENFELD, B. 1939 The specific nutritive requirements of *Clostridium acetobutylicum*. *Biochem. Jour.* 33: 1376-1389.
- (46) WEST, P. M. 1939 Excretion of thiamin and biotin by the roots of higher plants. *Nature* 144: 1050-1051.
- (47) WEST, P. M., AND LOCHHEAD, A. G. 1940 Qualitative studies of soil micro-organisms: IV. The rhizosphere in relation to the nutritive requirements of soil bacteria. *Canad. Jour. Res. (C)* 18: 129-135.
- (48) WEST, P. M., AND WILSON, P. W. 1939 Growth factor requirements of the root nodule bacteria. *Jour. Bact.* 37: 161-185.
- (49) WEST, P. M., AND WILSON, P. W. 1939 The relation of coenzyme R to biotin. *Science* 89: 607-608.
- (50) WEST, P. M., AND WILSON, P. W. 1940 Biotin as a growth stimulant for the root nodule bacteria. *Enzymologia* 8: 152-162.
- (51) WOOD, H. G., ANDERSON, A. A., AND WERKMAN, C. H. 1938 Nutrition of the propionic acid bacteria. *Jour. Bact.* 36: 201-214.
- (52) WOOLLEY, D. W., AND HUTCHINGS, B. L. 1939 Some growth factors for hemolytic streptococci. *Jour. Bact.* 38: 285-292.
- (53) WOOLLEY, D. W., AND HUTCHINGS, B. L. 1940 Synthetic media for the culture of certain hemolytic streptococci. *Jour. Bact.* 39: 287-296.

SURVIVAL OF BACTERIA ADDED TO SOIL AND THE RESULTANT MODIFICATION OF SOIL POPULATION¹

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It has long been recognized that in order to enable a new organism to become established in the soil, it is not sufficient to introduce large numbers of this organism: it is necessary also to modify soil conditions. A bacterium, fungus, or nematode added to the soil will become a member of the soil population only if the soil and environmental conditions are favorable for the growth of the new organism. This was found to hold true, for example, for the root-nodule bacteria when the specific host plant is present, for sulfur bacteria when sulfur is added to the soil, for mycorrhizal fungi in the presence of the specific host, and for insect-infecting bacteria or nematodes where the particular insect or its pupal stage is present in the soil.

To what extent this is true of other microorganisms is still a matter of speculation. The survival in the soil of the numerous bacteria added with stable manure has aroused great interest. The general conclusion seems to be now that these organisms survive only to a very limited extent. The answer to the question of the survival of all the organic matter decomposing bacteria, nitrogen-fixing bacteria, and many others added to the soil appears to be similar: in spite of the numerous claims to the contrary, beginning with the work of Caron (2) and Hiltner (6) and ending with that of Brown (1), Makrinov (10), Kostytshev (8), and many others, positive evidence is still lacking, except in very special cases (4). It has been shown, for example (11), that soils receiving stable manure contain larger numbers of thermophilic bacteria than do soils not so treated; the same is true of the occurrence of thermophilic actinomycetes (14).

The survival in soil of organisms (bacteria, fungi, actinomycetes) antagonistic to soil-borne plant diseases has also attracted considerable attention recently, the major difficulty being the establishment of the antagonist in the soil (5). Katznelson (7) has shown that the mere inoculation of microorganisms into a soil will lead to their rapid disappearance, as a result of a lack of a proper food supply, unfavorable environment such as reaction, competition for nutrients, or development of antagonistic soil organisms.

The belief in the role of stable manure as a soil inoculant persisted for some

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time (9), until Temple (13) and others succeeded in demonstrating that the multiplication of bacteria in soil as a result of addition of stable manure is due entirely to the organic constituents of the manure and not to its bacterial population, since sterile manure has the same effect upon bacterial multiplication as does fresh unsterile manure.

The disappearance of *E. coli* in soil was established, among others, by Skinner and Murray (12). This phenomenon was recently shown to be due to the development, in the soil, of active antagonists which destroy the *E. coli* (15). Dubos (3) has shown that the enrichment of soil with Gram-positive bacteria leads to the development of certain spore-forming bacteria which possess strong bactericidal properties against the former.

EXPERIMENTAL

The first experiment deals with a study of the influence of enrichment of soil with three bacteria, the last two of which are not indigenous to the soil; namely, *Aerobacter aerogenes*, *Escherichia coli*, and *Brucella abortus*. These organisms were grown on nutrient agar media, at 28° and 37°C. The cell material was collected, centrifuged, washed with sterile tap water, and again centrifuged. The washed cells were added to the soil and incubated at 28° and 37°C. A freshly drawn sample of Sassafra loam was used for this purpose. One thousand gram quantities of soil were placed in glazed porcelain pots and covered with glass plates. At intervals of about one week, fresh suspensions of the bacteria were added to the corresponding soils. Some of the soils also received 2 per cent CaCO₃ in order to neutralize the excess acidity.

Fresh lots of the suspension of the coliform bacteria were added as follows: one before the first count, five before the second count, and one before the third. Soil samples were taken, at various intervals, from several of the pots and were plated out on egg-albumin agar; the plates were incubated at 28° for 4 days.

The results presented in table 1 show that the coliform bacteria tended to disappear very rapidly from the soil. The total soil population, however, increased as a result of the addition of these bacteria. This bacterial multiplication may be due partly to the fact that the addition of bacterial suspensions to soil stimulated the development of certain specific soil organisms, principally of the antagonistic type. It is of particular interest to note here that spore-forming antagonists predominated in the soils enriched with *E. coli* and incubated at 28°C., but the soils enriched with *E. coli* and with *B. abortus* and incubated at 37°C. stimulated an extensive development of actinomycetes. The disappearance of *E. coli* in the soil was due not so much to the inability of this species to grow and multiply in the soil as to its destruction by soil antagonists. The fact that the increase in the soil population took place at the expense of the added bacterial suspensions is confirmed further by the formation of nitrate in the enriched soils (table 2).

The ability of coliform bacteria to survive and even to multiply in sterile

soil is shown in table 3. One hundred gram portions of moist sterile soil, untreated, treated with glucose (1 per cent), or treated with dried blood (0.5 per cent), were inoculated with *E. coli* and *A. aerogenes*, and incubated at

TABLE 1
Survival of bacteria introduced into soil and effect on soil population
Microorganisms in thousands per gram of soil

INCUBATION*	TEMPERATURE OF INCUBATION	INOCULUM	TOTAL ORGANISMS	COLIFORM BACTERIA
days	°C.			
5	28	Control	21,400	<200
5	28	<i>E. coli</i>	25,600	6,800
5	28	<i>E. coli</i> †	39,700	3,500
5	37	<i>E. coli</i>	22,800	4,700
33	28	Control	5,900	<10
33	28	<i>E. coli</i>	22,100	130
33	28	<i>E. coli</i> †	17,600	140
33	37	<i>E. coli</i>	23,000	10
37	28	Control	8,800	<10
37	28	<i>E. coli</i>	46,000	1,000
37	28	<i>E. coli</i> †	42,000	1,070
37	37	<i>E. coli</i>	36,300	2,900
120	28	Control	9,100
120	28	<i>E. coli</i>	40,000
120	28	<i>E. coli</i> †	36,300
52	37	<i>B. abortus</i>	60,200
82	37	<i>B. abortus</i>	212,500

* Elapsed time between last enrichment and plating was 5 days for the 5-day plating, 10 days for the 33-day plating, 4 days for the 37-day plating, 3 days for the 120-day plating, 10 days for the 52-day plating, and 2 days for the 82-day plating.

† Soil received CaCO₃.

TABLE 2
Influence of addition of E. coli upon nitrate formation in soil
Incubation—40 days

SOIL TREATMENT	TEMPERATURE OF INCUBATION	NO ₃ -N PER 100 GM. OF SOIL	pH OF SOIL
	°C.	mgm.	
Control.....	28	1.1	6.0
<i>E. coli</i>	28	2.6	5.5
CaCO ₃ + <i>E. coli</i>	28	4.2	7.5
<i>E. coli</i>	37	5.0	5.5

28°. After 10 and 26 days, the variously treated soils were plated on Endo medium and the number of viable cells determined. Both organisms were found to multiply rapidly in sterile soil, the addition of glucose having an unfavorable effect upon *E. coli* but not upon *A. aerogenes*. Dried blood had

little if any effect upon development of these two organisms. This tends to show that, as a result of heating the soil during sterilization, sufficient nutrients were liberated to enable the bacteria to make a good growth. The heating of soil with glucose seems to have resulted in the production of substances injurious to *E. coli*.

Several antagonists were isolated from the enriched soils by methods described elsewhere (15). In order to determine the effect of these organisms upon the microbiological population of the soil, two antagonistic bacteria, one

TABLE 3
Multiplication of coliform bacteria in sterile soil
Bacteria in thousands per gram of soil

INCUBATION days	E. COLI ENRICHED SOIL			A. AEROGENES ENRICHED SOIL		
	Soil alone	Soil + glucose	Soil + dried blood	Soil alone	Soil + glucose	Soil + dried blood
0	2.6	2.6	2.6	109	109	109
10	149,000	<1	70,000	48,000	1,660	77,000
26	138,000	<1	133,000	42,600	240,000	58,700

TABLE 4
Influence of products of antagonistic bacteria added to nutrient agar upon the development of a soil population
Bacteria in thousands per gram of soil

AGAR TREATMENT*	CONTROL SOIL	E. COLI ENRICHED SOIL
Water alone, 40 cc.....	1,510	17,700
Water, 35 cc., + filtrate of <i>Ps. aeruginosa</i> ,† 5 cc.....	570	10,700
Filtrate of <i>Ps. aeruginosa</i> , 40 cc.....	11	1,260
Water, 35 cc., + No. 58 filtrate,† 5 cc.....	1,050	16,400
No. 58 filtrate, 40 cc.....	400	6,370

* Additions to 40 cc. agar, double concentration.

† Filtrate = culture acidified (pH 4.5-5.0) and filtered through a Seitz; additions made to sterile agar.

very active, which proved to belong to the *Ps. aeruginosa* group, and the other less active, being a spore-forming soil bacterium (No. 58), were grown on a tryptone broth medium. The cultures were filtered through a sterile Seitz, and varying amounts of the filtrates were added to nutrient agar; this was prepared in double concentration to allow for its dilution by the filtrate. Two soils, one enriched with *E. coli* and one kept as control, were diluted with sterile water and different dilutions plated out by the use of the above agar. The plates were incubated for 7 days at 28°C., and the colonies were counted (table 4).

The filtrate from *Ps. aeruginosa* reduced to a considerable extent the number of soil microorganisms (except the fungi), even in very small concentrations; in higher concentrations, it greatly repressed the growth of both the bacteria and the actinomycetes in the soil; the fungi were not affected. The weaker antagonist (No. 58) had a more limited effect upon the development of the soil population on the plate. It is of further interest to note that, whereas the control plates had large numbers of spreading mucoid colonies, the plates receiving the filtrate from *Ps. aeruginosa* had only very small, many only pin-point, colonies.

TABLE 5

Influence of actinomycin upon the development of soil bacteria and actinomycetes on the agar plate

Microorganisms in thousands per gram of soil

ACTINOMYCIN PER LITER OF AGAR	CONTROL SOIL	E. COLI ENRICHED SOIL	B. ABORTUS ENRICHED SOIL
<i>mgm.</i>			
0	7,750	11,600	24,000
5	220	200	55
25	0	0	1

TABLE 6

Influence of actinomycin upon the microbiological population of the soil

Microorganisms in thousands per gram of soil

Incubation. .days	0		1		6		22	
Actinomycin added per 6 cc. of suspension	Nutrient agar	Egg- albumin agar	Nutrient agar	Egg- albumin agar	Nutrient agar	Egg- albumin agar	Nutrient agar	Egg- albumin agar
<i>mgm.</i>								
0	39	197	525	1,320	996	1,448	1,300	1,610
1.0	98	153	43	81	2,820	3,632*	7,590	7,300*
2.5	72	143	20	49	28	58	25	13.7
5.0†	50	117	7	40	8	28	1	1.7

* A pure culture of a small coccoid bacterium was obtained on egg-albumin plates of this treatment.

† Spore-formers predominated on plates from these treatments.

The foregoing experiment was repeated with a purified active substance isolated from a soil actinomycetes, designated as actinomycin (15). This substance was found to be highly bacteriostatic, inhibiting the growth of a large number of Gram-positive and Gram-negative bacteria and actinomycetes, but not of fungi. When added to the agar medium used for plating out soil suspensions, even in concentrations of 0.25 mgm. per 10 cc. of agar, it inhibited completely the development of all soil bacteria and actinomycetes capable of growing on the agar plate (table 5). The only organisms developing on the plates receiving the higher actinomycin concentration were some of the soil fungi.

In order to test the bactericidal effect of actinomycin upon the soil population, the following experiment was next made. A suspension of soil and water (1:1) was prepared, and 1-cc. portions were placed in a series of test tubes. An actinomycin solution (1 mgm. per 1 cc.) was added to each of the test tubes in amounts of 1.0, 2.5, and 5 mgm.; sterile water was added to bring the volume to 6 cc. After various periods of incubation at 28°C., the numbers of surviving organisms were determined by plating on nutrient and egg-albumin agars. The colonies were counted after 5 days' incubation.

The results (table 6) show that increasing concentrations of actinomycin added to a heavy suspension of soil in water had increasing bactericidal, as well as actinomycidal, effects upon the soil population. With the very low concentrations (1 mgm. per 6 cc. soil suspension), the reduction in the numbers of bacteria was followed by a rapid increase. The fact that, in this case, not the soil population as a whole but only certain bacterial types increased, points to the possibility that the low concentration of actinomycin had upon the soil an effect of partial sterilization: an initial reduction in bacterial numbers was followed by an increase. The effect of the higher concentrations of actinomycin was that of more complete, even if only selective, sterilization.

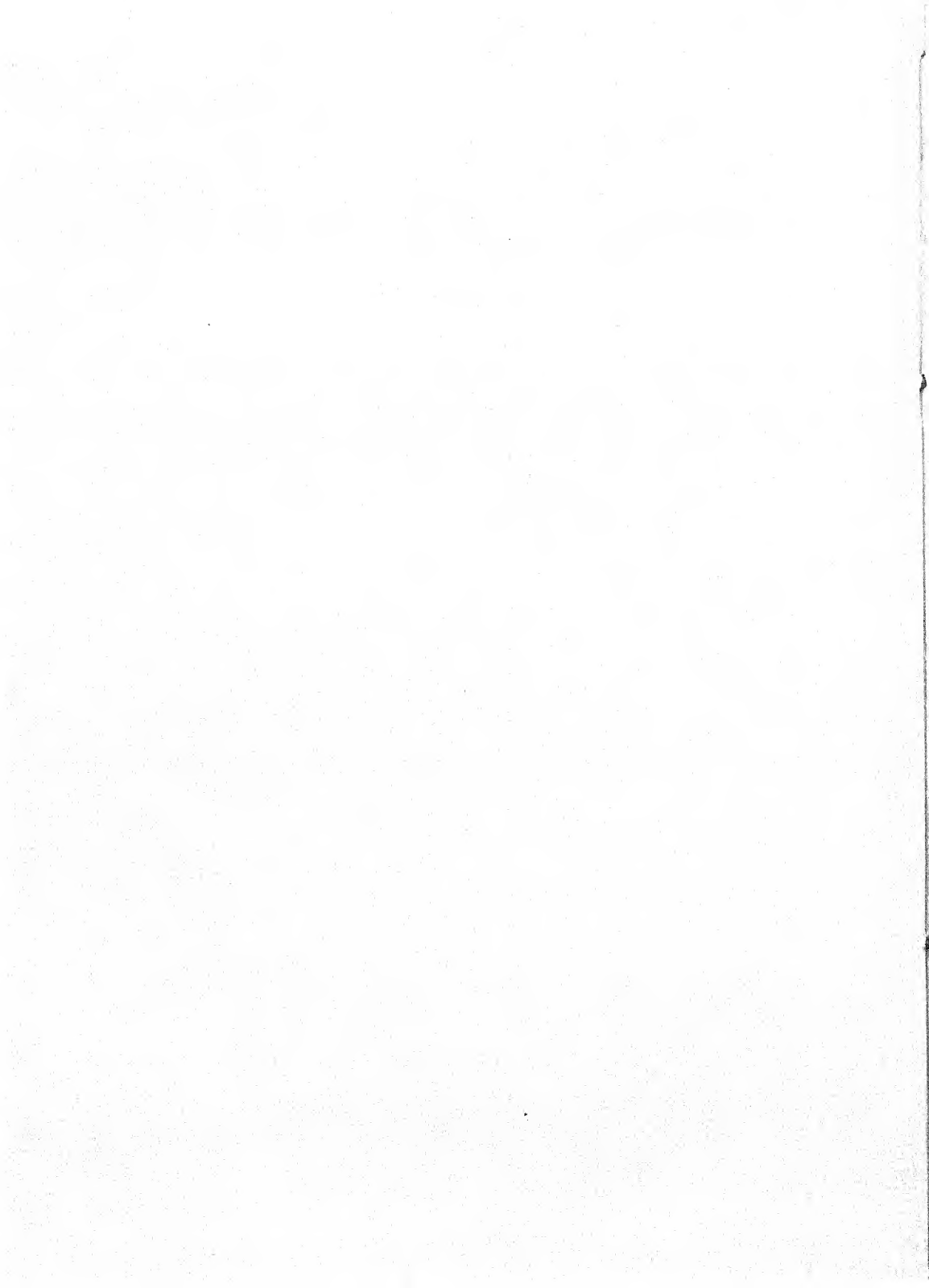
SUMMARY

A study of the survival of bacteria added to the soil but not indigenous to it, revealed the fact that these bacteria rapidly died out in the soil. This was accompanied by an increase in the numbers of soil bacteria and actinomycetes capable of developing on the plate. Further additions of cultures of bacteria led to their more rapid destruction. This was found to lead to the development in the soil of antagonistic organisms responsible for the destruction of the bacteria added. The nature of the antagonists depended upon the bacteria added, the soil treatment, and the temperature of incubation of soil. Several antagonists were isolated from the soil, and their effect upon various bacteria as well as upon the total soil population was studied. It was shown that sterile filtrates of these antagonists can reduce considerably the numbers of bacteria and actinomycetes in the soil. The fungi were least affected, if at all. The most active bacteriostatic and bactericidal substance was obtained from a soil actinomycetes.

REFERENCES

- (1) BROWN, P. E., AND HART, W. J. 1925 Soil inoculation with *Azotobacter*. *Jour. Amer. Soc. Agron.* 17: 456-473.
- (2) CARON, A. 1895 Landwirtschaftlich-bakteriologische Probleme. *Landw. Vers. Sta.* 45: 401-418.
- (3) DUBOS, R. 1939 Bactericidal effect of a soil bacillus on gram-positive cocci. *Proc. Soc. Exp. Biol. and Med.* 40: 311-312.
- (4) GAINES, P. L. 1925 Inoculating soil with *Azotobacter*. *Soil Sci.* 20: 73-86.
- (5) GARRETT, S. D. 1939 Soil-borne fungi and the control of root-disease. *Imp. Bur. Soil Sci. Tech. Commun.* 38.

- (6) HILTNER, L. 1921 Über die Impfung der Futter und Zuckerrüben. *Mitt. Deut. Landw. Gesell.* 26: 243.
- (7) KATZNELSON, H. 1940 Survival of microorganisms introduced into soil. *Soil Sci.* 49: 283-293.
- (8) KOSTYTCHEV, S. P. 1930 Conclusions of agro-chemical character based upon the studies in soil biodynamics. *Bul. Inst. Agr. Microbiol. U. S. S. R.* 4: 29-40.
- (9) LIPMAN, J. G., ET AL. 1914 The influence of bacteria supplied in manure, on the decomposition of green manure (legume and non-legume). *N. J. Agr. Exp. Sta. Bul.* 268: 22-25.
- (10) MAKRINOV, I. A. 1924 Is it possible to make a bacterial soil preparation for non-legume crops? *Soil Sci.* 17: 31-38.
- (11) MISCHUSTIN, E. N. 1935 Thermophilic bacteria as an indicator of state of soil cultivation. *Microbiol. Lab. V IUAA, U. S. S. R.*, p. 55-80.
- (12) SKINNER, C. E., AND MURRAY, T. J. 1926 The viability of *B. coli* and *B. aerogenes* in soil. *Jour. Infect. Diseases* 38: 37-41.
- (13) TEMPLE, J. 1911-12 The influence of stall manure upon the bacterial flora of the soil. *Ga. Agr. Exp. Sta. Bul.* 95; *Centbl. Bakt.* (II) 34: 204-223.
- (14) WAKSMAN, S. A., UMBREIT, W. W., CORDON, T. C. 1939 Thermophilic actinomycetes and fungi in soils and in composts. *Soil Sci.* 47: 37-61.
- (15) WAKSMAN, S. A., AND WOODRUFF, H. B. 1940 The soil as a source of microorganisms antagonistic to disease-producing bacteria. *Jour. Bact.* 40: 581-600.



A NEW SPECIES OF SULFUR-OXIDIZING BACTERIA FROM A COPROLITE

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Continuing the senior author's investigations of the last dozen years or so on the survival of living bacteria in ancient materials, we had occasion to use coprolites of the Triassic. In testing material of the interior of these coprolites, growth of a variety of organisms, usually of several forms, was obtained in some of the media used. Among these was an alkaline sulfur medium prepared after Jacobsen's formula. In this at least one organism was found with a definite power to produce sulfates from sulfur and from thio-sulfate. Sulfate production by this organism was more rapid and marked in thiosulfate than in sulfur media, but in both it was very definite. A study of the organism revealed that it possesses characteristics quite distinct from those of other sulfate-producing organisms, and it is, therefore, recognized as a new species. Though it is not yet certain that the organism belongs to the genus *Thiobacillus*, the presumption is in favor of classifying it there. It is thought opportune now to describe this organism and to offer the evidence for giving it a specific status new to science.

The organism was first discovered in the crude cultures of the coprolite material as an exceedingly thin rod about 0.1 to 0.2 μ in width and about 6 to 8 μ in length. The occurrence in these same cultures of S-shaped rods and vibrios in goodly numbers led us at first to think of it as a spirillum, but repeated culturing, resulting in its isolation in pure culture, and perhaps its most frequent occurrence in straight-rod form lead us now to think of it as a bacillus rather than as either a spirillum or a vibrio. That it is probably not a spirillum is indicated by the fact that in establishing its motility we found that it possesses only a single polar flagellum. Regarding its possible character as a vibrio, we shall bring forward discussion below leading to a negative conclusion. Continued culturing of the organism in pure form revealed the fact that it is exceedingly variable in length. Though the mode is 6 to 8 μ , the range may be from 3 to more than 40 μ in filamentous form.

¹ Our thanks are due to C. L. Camp, of the University of California, for furnishing the coprolite in which we discovered the organism described in this paper; to Charles Gilmore, of the Smithsonian Institution, for furnishing other materials; and to C. B. Van Niel, of the Hopkins Marine Station, for advice relative to the systematic position of the organism.

The description of the organism may be summarized as follows:

Very thin long rods 0.1 to 0.2 by 6 to more than 40 μ .

Motile with a single polar flagellum. Occasionally occurs as spirillum and in large numbers as vibrio.

Slight but very definite growth on peptone soil extract agar.

Difficult if not impossible to induce growth in nutrient broth.

Thiosulfate agar plate: Slow but good growth as tiny watery colonies raised above surface of medium. In one case only, colonies were observed to become white from precipitated sulfur.

Thiosulfate broth: Little or no turbidity. No pellicle. No sediment. Reaction changes from pH 7.6 to 6.1.

Sulfur solution medium (alkaline): Good growth. No turbidity. Reaction about pH 7.8.

Autotrophic facultative.

Aerobic.

Distinctive characters: Unusual morphology. Oxidizes thiosulfate and sulfur to sulfate. Probably no intermediate tetrathionate step in thiosulfate oxidation. Prefers alkaline reaction but has a pH range from 4.6 to 10.

Source: Coprolite from Triassic of Arizona.

In comparing this organism with those described by Starkey² in 1935 and others mentioned in Bergey's *Manual of Determinative Bacteriology*³ we find that its characters do not fit any of their descriptions. The extraordinary length of the filaments at the extreme, the impressive length of the rods or vibrios at the mode, and the not inconsiderable length of the smallest rods place this organism in a class by itself morphologically. Its thinness is certainly extraordinary. Delicacy in focusing with the oil immersion objective is essential to give a clear view of the organism, because its thinness brings it close to the point of resolution of our lenses. The longest of all the organisms in the genus *Thiobacillus*, according to the sources just quoted, is *T. thermitanus* (Emoto) which is reported to have a maximum length of 5 μ . When we compare this with the dominant length of our organism, which exceeds this maximum, and bear in mind what we have just said about its other morphological characters, it becomes clear that, from that point of view alone, we are dealing with a new species. But there are physiological characters of our organism which are also distinct from those of other species in this group. The group of thermophilic forms described by Emoto in this genus can all withstand much more acidity than can our organism. They are reported incapable, moreover, of living on organic media, but our organism grows on peptone soil extract even though it does not seem to tolerate nutrient broth. By reason of its tolerance of at least one organic medium and its definite and fairly efficient oxidation of sulfur, our organism stands in contrast to the organisms studied by Starkey, for *T. trautweinii* or its related form either does not oxidize sulfur at all or does it exceedingly ineffectively, and *T. thioparus*,

² Starkey, R. L. 1935. Isolation of some bacteria which oxidize thiosulfate. *Soil Sci.* 39: 197-221.

³ Bergey's *Manual of Determinative Bacteriology*, ed. 5. 1939.

though it oxidizes sulfur very slowly, does not tolerate organic media and besides is morphologically entirely different from our organism. In another respect physiologically our organism stands in sharp contrast to all the organisms mentioned: all the members of the genus thus far described are reported as rendering turbid thiosulfate or sulfur media or both and usually very quickly; our organism fails to induce such turbidity.

Some interesting aspects of the organism under consideration present themselves in connection with the study of its motility in comparison with that of other organisms in the genus *Thiobacillus*. Our organism has a single flagellum, as is shown in plate 1, figure 5. The flagellum is never as long as the organism itself but is more likely to be one-third to one-half the length of the bacillus. When we compare this with the organisms described by Starkey, we find that his organism A, which he named *T. novellus*, is not motile at all. Starkey's organism B, which he thinks is probably identical with *trautweinii*, is motile, but it has several peritrichic flagellae. Starkey's organism C is again like his organism A—nonmotile. With respect to motility, therefore, there is no correspondence between any of the organisms described by Starkey and our organism. On the other hand, all of the organisms in this genus described by Emoto are motile and have only a single flagellum like our organism, but Emoto's organisms frequently have flagellae which are very much longer than the organisms.

One or two other points with respect to the studies under consideration here require special mention. The first concerns the deeply stained, coccuslike bodies which are frequently found in the cultures of our organism, as shown in plate 1; and the second relates to the frequency of occurrence of vibrio forms and even spirillum forms of the organism.

It is, of course, easy to think of the deeply stained coccuslike bodies as possible spores of the organism, but we have grown so habituated in bacteriology to think of spores as bodies which do not take stains solidly except by special staining methods that it may seem for the time being questionable whether the bodies referred to are spores. Having often seen bodies that are definitely spores in other organisms take the stain solidly, we are not sure about the validity of the criterion with respect to the staining possibilities of spores. Nevertheless, assuming, as indicated, that we think that criterion valid, then it may still be possible that the deeply stained bodies which we have shown as belonging to our organism may be regenerative bodies. Repeated observations which lend color to this hypothesis or to the spore hypothesis for these somewhat mysterious bodies show that frequently one of the extremely thin rods or vibrios which characterize our organism is seen to be attached to and to issue from one of the deeply stained bodies, which may be regarded as regenerative.

The frequency with which the vibrio form especially or the spirillum form is encountered in our cultures of the new sulfur-oxidizing organism is not inconsistent with the view that our organism belongs to *Thiobacillus* and may be

primarily regarded as a bacillus. It is not unlikely that other members of the genus *Thiobacillus* might also be found to appear as vibrio forms if they were not so short; in fact, photographs accompanying descriptions of other members of the genus *Thiobacillus* show occasional vibrios even among the short rods. As our organism is much longer than the other members of this genus, there is more opportunity for curvature of the rods, or at least for greater chance to observe the curvature, than in the shorter organisms. Similar comments would probably apply to the spirillum forms which are occasionally encountered and to the especially interesting wavy forms of the filamentous type, which are shown in plate 1, figure 4. Whether this view of the situation is correct or not, the vibrio forms of our organism, the spirillum, and the somewhat extraordinary wavy forms of the filamentous rods are not uncommon.

In view of the characteristics which differentiate our organism from related forms, we propose for it the status of a new species under the name *Thiobacillus coproliticus*.

SUMMARY

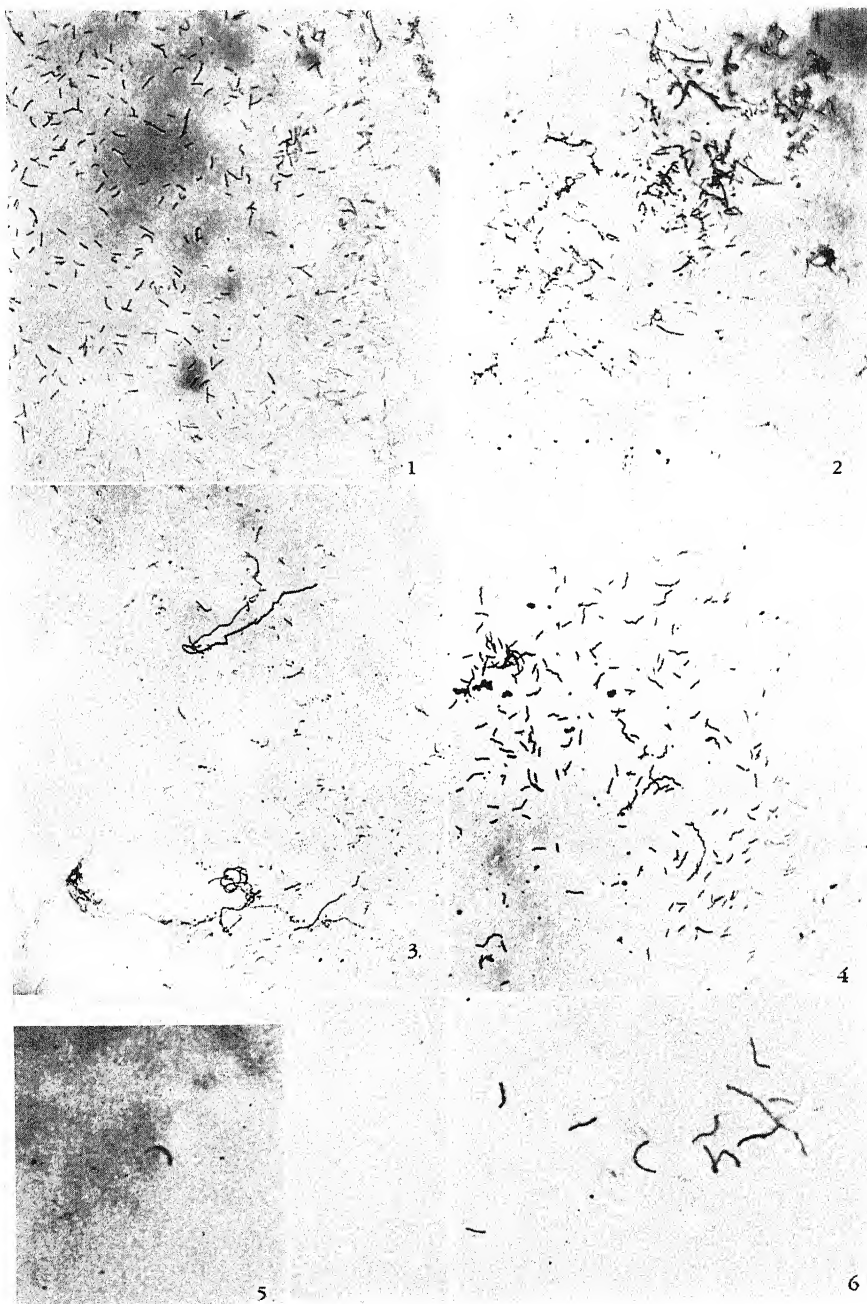
A new organism, probably belonging to the genus *Thiobacillus*, is described, which has the power of oxidizing sulfur, as well as thiosulfate, in inorganic media. The characteristics of this organism are described, and its differences from other forms in the genus are given. Since the organism does not correspond morphologically or physiologically to any of the known forms in the genus, we regard it as a new species which we name *Thiobacillus coproliticus*.

PLATE 1

A NEW SPECIES OF SULFUR-OXIDIZING BACTERIA, *THIOBACILLUS COPROLITICUS*

- FIG. 1. Culture about 2 weeks old in thiosulfate medium. $\times 480$.
FIG. 2. Older culture from sulfur medium. Note regenerative bodies. $\times 480$.
FIG. 3. Old culture in thiosulfate medium. Note filamentous forms. $\times 480$.
FIG. 4. Wavy forms of rods and filaments in peptone soil extract medium. Note occasional rods issuing from regenerative bodies. $\times 480$.
FIG. 5. Vibrio with flagellum. $\times 900$.
FIG. 6. Two more rods with flagellae. $\times 900$.

C. B. LIPMAN AND E. MCLEES



FIGS. 1-6

SOIL AND WATER CONSERVATION IN THE SOUTHERN GREAT PLAINS

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Large areas in the Great Plains were ravaged during the period 1933 to 1936 by dust storms of an intensity and scope unparalleled in American history. Soil from the farm lands of Kansas, Colorado, New Mexico, the Texas and Oklahoma Panhandles, and other parts of the 350 million acres comprised in the Great Plains was blown nearly two-thirds of the distance across the continent out over the Atlantic Ocean. Thousands of fields were swept of fertile topsoil down to plow depth and others were covered with hummocky drifts of wind-driven soil or dunes of wind-assorted sand. Whole counties had experienced complete crop failure; hundreds of families had left their farms and homes to look for new opportunity somewhere on the unknown horizons of the nation.

In those days, there were many who frankly believed that the Great Plains was through as an important agricultural region. Such meaningless appraisals as "that country is no good anyway; why not get out of it and forget it; give it back to the Indians" were frequently heard.

Today the outlook, generally, is better than it has been in years. Because of the increasing use of water-conserving farm measures, the situation has changed to a promising outlook in many localities where a little while ago conditions were essentially intolerable and the future was very dark. Over most of the region, dust storms have been declining in frequency and severity steadily since 1936. Thousands of acres of severe blow land have been stabilized under a permanent cover of grass; hundreds of farms have been fully treated for soil and water conservation. Fields that were completely barren and wind-swept three or four years ago are now covered with good crops of wheat and sorghum.

Probably the most encouraging development of the last three years in the Plains country has been the dissipation of many former doubts and fears. The cooperative accomplishments of farmers and technical men in Soil Conservation Service work areas have proved once and for all, I think, that soil erosion *can* be controlled in this region if farmers have the will, the ingenuity, the energy, and the necessary knowledge to carry out the job. And the recent rapid formation of soil conservation districts by farmers under state laws is a clear indication that the people of this region are prepared to fight to save their land.

PERMANENT CONTROL OF EROSION

At the present time we know much more about controlling soil erosion in this region than ever before. We know more about soil-saving tillage methods and cropping systems; more about conservation of rainfall, establishment of grass on badly eroded land, control of shifting sand dunes, and many other phases of the problem. The essential elements for a broad program of soil and water conservation in the Great Plains can now be stated definitely. Special problems calling for additional information still exist, but specialists are striving diligently to find the answers.

Specifically, permanent control of erosion in this region involves: conservation and more efficient use of rainfall; establishment of a permanent vegetative cover on the severely eroding and erodible lands; the adoption of cropping systems based on soil and moisture conditions at planting time; adaptation of cropping systems and cultural practices to soil and topographic conditions; the protection and efficient utilization of crop residues; the use of protective vegetated strips and borders of grass, shrubs, or trees; the proper distribution and regulation of grazing on range lands; stabilization of moving sand dunes on both farm and range areas; adjustment in size of farms; and purchase of submarginal areas in order to make needed land use and population adjustments through the development of idle land for productive use and through the installation of soil and water conservation practices.

The principal objective of this paper is to define briefly each of the separate elements of such a broad program of soil and water conservation and to present some of the more recent discoveries and accomplishments.

CONSERVATION AND UTILIZATION OF RAINFALL

As those who are acquainted with the Plains know, water is and always has been the key to successful agriculture in this part of the country; it is also the key to soil conservation. Without water in the soil, there can be no vegetative cover to anchor the land against the whirl of wind and the rush of rainwater. Without water, cultivated soil, depleted of binding grass roots and spongy organic matter, is turned into a dry, powdery substance that takes off with the slightest breeze and may be blown over hundreds or even thousands of miles. Without conservation and careful utilization of the rainfall, there can be no lasting security in this region for people on the farm.

In the last two generations there were various proposals for increasing rainfall over the Great Plains region. During the homestead period, for example, the belief that "rainfall follows the plow" was widely held. The answer to the drought problem of the Plains, many observers felt, lay in a constant enlargement of the cultivated acreage, a constant stripping away of the native vegetative cover. Another school of thought held that trees on the Plains would somehow insure more dependable rainfall; and the Timber Culture Act of 1873 was passed partly because of this hope. But sooner or later these schemes and others proved futile. Recent climatic research indicates with some

finality that neither ground cover nor lack of it has any appreciable effect on the amount and distribution of rainfall on the Plains. In fact, there is no known practical method for increasing the rainfall of any region; and there seems little prospect that any such method will be discovered in the near future.

In planning for soil and water conservation in the Great Plains, therefore, we simply have to deal with the stubborn fact that this is and probably will continue to be a region of low rainfall and recurrent drought. The only sensible course is to adjust agriculture to the realities of a climate that is occasionally harsh and, through practical measures of conservation and utilization, to increase the efficiency of the rain that does fall. Past and present land problems in this region can be traced in part to drought but also in a large measure to the fact that *too much of the available rainfall has been allowed to go to waste.*

During the last few years, however, a great deal of encouraging progress has been made in this region toward more efficient water conservation and water utilization. In Soil Conservation Service work areas, contour tillage, level terracing, strip cropping, and basin listing have helped to hold rainfall on the land. Cover crops and crop residues have served to keep the ground surface open, absorptive, and resistant to erosion. Spreader structures of many kinds have taken water that formerly was running to waste—in roadside ditches, for example—and distributed it over cultivated fields and grazing lands. In fact, greater advances have recently been made in the utilization of rainfall in this region than anyone could have foreseen just a few years ago. Thus the plainsman, like the farmer of the humid region, is finally coming to the realization that it is really practicable to alter the climate, in the sense of its relation to plant life, through the simple expedient of saving that part of the rainfall which has been running to waste and storing it in the reservoir of the soil for the benefit of plants.

Tests made in the erosion control demonstration area in Seward County, Kansas, indicate the kind of results that have been obtained from practical water conservation work. For several years, wheat farmers have been practicing summer fallow as a means of storing moisture in the soil for succeeding crops of winter wheat. Before the erosion control demonstration started in the fall of 1935, preparation for fallow consisted of plowing straight furrows, often up and down the slope. More recently, however, a large number of farmers in the county have been plowing across the slope on the contour and have built terraces to increase the efficiency of their summer fallow.

During the fallow period from April to September, 1938, inclusive, 14.79 inches of rain fell in the project area; but during the wheat-growing season from October to the following June, inclusive, the precipitation was only 6.64 inches. Moisture tests and wheat yield records were taken in October on 989 acres of land that was contour-fallowed and protected with terraces. The contour-tilled fields showed an average of 16.78 per cent moisture in the soil as against an average of 14.42 per cent on 1,180 acres where straight tillage fallow was

practiced. The moisture content at wilting stage for wheat in the Liberal project is 11.84 per cent. Accordingly, this difference—an increase in available moisture of about 91 per cent, or 4.94 against 2.58 per cent of moisture in the soil—takes on added meaning when translated into bushels of wheat. The average yield on the straight-tilled fallow fields was 7.86 bushels an acre, whereas contour-fallowing brought an average yield of 12.77 bushels—a gain of nearly 5 bushels an acre, or 62 per cent. This amounted to an increased production valued at \$4 an acre above similar untreated land.

These tests show clearly that, by means of contour tillage and terracing, Plains farmers can, under ordinary conditions, be almost certain of a fair wheat crop even with very light rainfall during the growing season.

Examples of this kind could be multiplied throughout the Great Plains. Near Clovis, New Mexico, for example, wheat yields on 30 terraced farms were compared last year with those on 34 comparable farms where there was no terracing or contour tillage. The terraced farms produced 3.36 bushels more per acre on the average, with the result that the farmers who had terraces realized about \$2 more an acre on the year's work than did their neighbors. The increased crop production in that one year more than paid for the costs of terrace construction; and the terraces, if properly maintained, will go on producing dividends and providing soil protection for many years to come.

Even more striking are the results obtained during the last two years near Hereford, Texas, on two farms that lie on opposite sides of a road. Both farms have approximately the same kind of soil and slope; both farmers have planted the same kind of wheat at the same time of year. But while one man has farmed his fields in the customary way, his neighbor across the road has built terraces and plowed on the contour to hold back the rainfall. In fact, this conservation farmer has even diverted water from the roadside ditch and spread it over his fields to help the growth of wheat. Last year, with heavier than average rainfall, the yield on the contoured and terraced land was 36 bushels to the acre, and on the other farm it was only 15 bushels. In 1938, when rainfall was not so high, the conservation farm produced 23 bushels to the acre against only $4\frac{1}{2}$ bushels—scarcely enough to harvest—on the nonconservation farm across the road. In other words, the benefits of water conservation work in this region appear to be good in times of low as well as high rainfall.

ESTABLISHING A VEGETATIVE COVER

The job of establishing a protective cover of vegetation on lands that constitute a blow hazard is complicated in the Plains country not only by the limited rainfall but by a shortage of harvested native grass seed. Recently, however, considerable progress has been made in overcoming both of these difficulties. Contour-furrowing land before it is seeded to grass or some other protective cover has proved helpful in preventing failure by storing up the rainfall and delivering it to the plant roots, and new methods of harvesting

grass seed have been especially encouraging. Formerly, hand strippers were used almost exclusively by the Soil Conservation Service in collecting Plains grass seed not otherwise obtainable in quantity. The results were only moderately satisfactory. In August, 1938, a rancher near Hereford, Texas, tried a 12-foot combine on a section of blue grama grass that had produced an abundance of seed. Using this method, the rancher collected a much higher quality of seed than had ever been obtained by the hand-stripping practice. As a result, the Service in September of the same year tried a 5-foot standard combine in collecting side-oats grama seed in the same general area. This trial proved definitely that large collections of seed could be made economically with regular commercial types of combines. Since that time, combines have been used more and more extensively by the Service in its seed harvesting. Last year, collections of important species of grass in the Southern Plains region alone totalled 198,000 pounds—considerably more than the harvest for any previous year.

The seed collected in this way have been used to tie down thousands of acres of blow land in the Great Plains. But grass is only one of the weapons used. Before native grass can be grown successfully on most of the blow land of the Southern Plains the soil blowing must be checked. Usually the land is stabilized by the establishment of a dense cover of sorghum prior to grass planting.

In the West Baca Soil Erosion District, broom corn and cane have been used to stabilize blowing soil in one of the most extensive revegetation jobs ever attempted in the Southern Great Plains. This work was started in the fall of 1938, when farmers in the western part of Baca County banded together and formed a soil conservation district under the recently enacted Colorado law. Realizing that abandoned blowing land was a definite hazard to all adjoining fields, the members of the district pooled their resources, leased 11,000 acres of this blow land, and listed it as an emergency measure. Last year the district leased 56,000 acres of abandoned land, leveled it off with heavy drags, and planted it to broom corn and other erosion-resistant sorghums. At the same time, the Soil Conservation Service established a soil-protecting cover on about 14,000 acres of government-owned land in the county—land that had been hard used by the wind. This year Baca County, once one of the most critical erosion problem areas in the entire Great Plains, has had fewer dust storms than at any other time since 1933. The thousands of acres of stabilized land have had a great deal to do with this. And the land retirement and stabilization work of the district is still going forward.

CROPPING SYSTEMS AND SOIL CONDITIONS

Over the Great Plains as a whole, hundreds of fields that should be under grass are in cultivation. But it is a serious mistake to consider regrassing the whole answer to the problems of the Plains. Actually, it is only the more sandy and shallow soils, generally, that require a permanent cover of vegetation. On the majority of farms crops can be grown on the more productive,

less erodible lands of relatively high clay content ("tight land") without excessive danger to the soil, provided proper precautions are taken. Most of the trouble comes when grain is seeded in dry soil, when so often crops fail and the land is left without any vegetative protection against the wind, such as even a crop stubble affords.

Tests made by the Kansas Agricultural Experiment Station in recent years show that under Great Plains conditions, the chances of producing a successful crop of grain are almost directly proportionate to the amount of water in the soil at planting time. Reporting on these experiments, R. I. Throckmorton, head of the department of agronomy, Kansas Agricultural College, summarized the results as follows:¹

... seeded in dry or nearly dry soil, the crop (wheat) failed 71 percent of the time, ... seeded in soil with one foot of moisture the crop failed 34 percent of the time or about one chance out of three for failure.

When moisture had penetrated to a depth of two feet at seeding time, there was only about one chance in seven of crop failure ... three chances out of five of obtaining a wheat yield of ten bushels or more per acre ... three chances in ten of obtaining a yield of twenty bushels or more per acre.

When the soil was wet to a depth of three feet or more, there was only one chance in ten of a crop failure.

In erosion control demonstration areas, grain farmers cooperating with the Soil Conservation Service are following a flexible system of cropping based on this highly significant relationship between crop yields and moisture in the soil at seeding time. They plant wheat only when moisture conditions are right—when there is enough water in the soil for reasonable assurance of a satisfactory crop. When soil moisture is less than 2 feet deep at planting time, they ordinarily depend on stubble left from previous crops or apply emergency treatment to protect the land through the winter and plant sorghums or some other drought-resistant crop in the spring. This system has a twofold advantage: it safeguards the farmer against needless crop losses and safeguards the land against unnecessary wind erosion.

Getting the maximum benefit from available rainfall in the Plains country, however, is not simply a matter of holding the rainfall on the land. In this region of hot, dry summer winds, there is also an extremely serious problem of evaporation. Even where there is no runoff, this factor sometimes accounts for a large percentage of the available rainfall and has a definite adverse effect on subsequent crop yields. In this connection, recent experiments conducted by the Soil Conservation Service in cooperation with the Nebraska Agricultural Experiment Station have yielded some interesting and highly significant results.

In the spring of 1938, seven experimental plots were laid out on the agronomy farm near Lincoln, Nebraska, and subjected to different kinds of treatment for the purpose of storing up moisture during the summer fallow period. One of the plots was simply plowed, another was disked, and a third was basin-

¹ *Country Gentleman*, March, 1936.

listed. On each of the four other plots, 2 tons per acre of wheat straw were applied to the land in different ways: on one, decayed straw was plowed in; on another, fresh straw was plowed in; on a third, fresh straw was disked; and on the fourth, it was spread over the ground and kept on or near the surface by a system of subsurface tillage which only partially worked the straw into the soil. An eighth plot, kept bare by removal of vegetation, was untreated. During the summer fallow period, April 23 to September 8, 1938, 17.9 inches of rain fell on all the plots. The amounts of water saved, in terms of surface inches and of percentage, are shown in table 1.

Where straw was applied on the surface, 54.3 per cent or 9.72 inches of the water was saved. This is more than 2 inches greater than the amount conserved under any other kind of treatment and better than two and a half times

TABLE 1

*Effect of straw and different tillage treatments on the storage of water in the soil near
Lincoln, Nebraska*

PLOT NUMBER	TREATMENT	PART OF 17.9 INCHES OF RAINFALL CONSERVED		DEPTH OF WATER PENETRATION
		<i>surface inches</i>	<i>per cent</i>	
1	Straw, 2 tons disked in	6.92	38.7	5
2	Land disked, no straw*	3.49	19.5	4
3	Straw, 2 tons on the surface	9.72	54.3	6
4	Straw, 2 tons plowed in	6.12	34.2	5
5	Land plowed, no straw*	3.71	20.7	4
6	Decayed straw, 2 tons plowed in	3.12	17.4	4
7	Basin listed	4.95	27.7	5
8	Bare unplowed†	1.23	6.87	2

* Either plot 2 or 5 could be used as check, since both were kept free of vegetation by the usual practice of summer fallow. Probably plot 5 is preferable from the standpoint of farm practice.

† Kept bare by taking off vegetation. This is not a farm practice and is only of technical interest.

that on plowed land. Moreover, the amount of water saved on the land plowed without straw is by no means low. It is in line with other summer fallow experiments that have been conducted throughout the Great Plains region. Consequently, these experiments indicate plainly that this kind of utilization of crop residues *increases* the efficiency of water conservation by a highly significant margin. The method is a phase of stubble-mulching.

The results with basin listing at the same location are of particular interest because on land treated in this way there was virtually no runoff, yet the amount of water conserved during the season was only half the amount stored under straw. This emphasizes the fact that prevention of runoff is not the full solution of the moisture problem on the Plains. On the basin-listed plot the losses due to evaporation from the convoluted bare surface tended to offset the gains due to prevention of runoff. The basin listing did prevent water

erosion, but this was also true where the straw treatments were used. And the straw, by acting as a kind of mulch, had the important added advantage of reducing evaporation losses.

Similar experiments have also been conducted at the same station in preparing a seedbed for wheat and in the conservation of moisture on corn land. In all cases, the value of leaving crop residues on the ground surface has been clearly apparent. Used in this way, as a "mulch," the residues produce three distinct benefits: first, they check runoff by keeping the surface soil open through the surface protection afforded by the vegetation residues, so that nearly all the water soaks into the soil; second, they reduce evaporation from the ground surface; and third, they help to protect the land against both wind and water erosion. Since the application of crop residues as a mulch is a comparatively simple practice, involving almost no cash outlay, the possibilities for its use in the Plains country and humid regions also are far-reaching. In fact, it looks like one of the most promising of the many methods that have recently been proposed as an antidote for low rainfall in this region.

In a letter to the writer, August 10, 1940, Dr. Duley² says:

The major problem of "subsurface tillage," or "crop residue culture," is the mechanical one of working beneath the surface and leaving all the residues on top, then seeding or planting through these residues, and later, in the case of row crop, cultivating beneath the surface.

We are succeeding eminently in this, not only in the simple practice of summer fallow, but with every other phase of management.

We view the use of outside sources of mulch material such as straw from stacks as a very minor phase of "subsurface tillage" practice, to be applied only in critical erosive spots where the normal residues are inadequate to meet the situation.

The idea is rapidly sweeping our section of the country and several hundred farmers are putting it into practice in one way or another.

In the Southern Great Plains utilization of standing stubbles by maintaining them in fields as long as practicable has been one of the important features of the regional soil conservation program since its inception in 1934. The practice has been of great help in establishing resistance to soil blowing.

Last year, the writer visited a farm near Gainesville, Georgia, where for several years the owner, Mr. M. E. Gowder, has been practicing a type of agriculture that conforms with the findings of the research work referred to above. He operates a small farm in a part of the hilly Piedmont section where erosion has been terribly destructive. Few farms are unaffected. Over large districts virtually every cultivated acre has been affected, and tens of thousands of acres have been made unfit for cultivation.

Mr. Gowder never plows under crop residues; rather, he plows through them so as to leave as much as possible of all stubble—corn, cotton, cowpeas, or whatever he grows. Even when his land was cleared, the leaves and twigs of the forest floor were plowed through, not under. In addition to preserving

² The experiments referred to are being carried on cooperatively by F. L. Duley, of the Soil Conservation Service, and J. C. Russel, of the University of Nebraska.

crop residues, Mr. Gowder uses terraces, cultivates on the contour, and plows with a narrow "scooter" plow to a depth of about a foot; as a result, his soil has practically no erosion and at the same time conserves much more of the rainfall than does his neighbors'. He makes much better than the average yields of the locality and is reported to be farming at a profit while keeping his children in school and his farm plant completely intact.

This kind of conservation farming is needed on millions of acres. It is one of the measures of land protection that is included in the program of the Soil Conservation Service. Far too much of the erodible fields of America is kept bare of protective cover over too much of the year.

ESTABLISHING WINDBREAKS

Another practice that has proved valuable in reducing evaporation losses and in checking wind erosion is the establishment of windbreaks of trees or hedges along the borders of cultivated fields and along strategic lines through fields. Trees, of course, are not a natural growth over most of the Great Plains country. But adaptable species can be made to grow in this region, particularly in the more favorable situations, provided enough care is taken in the preparation of the sites and provided also the windbreaks are properly maintained. Once established, a good field windbreak is a decided asset to the farmer. The trees help to control wind erosion by diverting air currents upward and by breaking up the sweep of low-moving winds. At the same time, they seem often to have some effect on moisture conservation in strips along the lee side of the windbreaks.

GRAZING ON RANGE LANDS

On many range areas in the Great Plains region, the original grass cover has been seriously weakened in recent years by the combined effects of overgrazing and drought. Undesirable plants have tended to replace the more useful grasses, and in some areas the land has been left bare. As a result, erosion by both wind and water has been extremely severe, and many ranchers have found it increasingly hard to provide their livestock with adequate forage.

Where soil and water conservation work has been carried out on range lands, improvement in the grass cover has been especially marked since 1935 and 1936, not only in the Plains but in grazing areas farther west. A good example is a block of 112 thousand acres in central New Mexico, acquired by the government under the land utilization program, where the grass was thinning out badly only a few years ago. Since that time much helpful conservation work has been done on this tract. Stockmen have adjusted the size of their flocks and herds to the actual carrying capacity of the range. New fences have served to keep livestock out of sparsely vegetated areas until these have had opportunity to recover; new watering places have helped to bring about a more uniform distribution of grazing. Contour furrows and water-

spreading installations or measures have played an important part in saving the rainfall and feeding it to the grass roots. As a result of all this work, the grass on this particular range is better than it has been for years, in spite of the record drought of 1939.

STABILIZATION OF DUNES

Since 1934, countless small and large dunes have formed on the Great Plains. Some are scattered and incidental; others exceed 1,000 acres in extent and 10 feet in height, dominating the landscape of wide areas. Their spread suggests conditions which prevail in parts of the Sahara border, as J. R. Ainslie³ describes them:

A few months ago I had the opportunity of visiting the French Niger Colony lying to the North of the Nigeria boundary; that country is very largely desert and includes within its area probably the most dreaded desert region in the world; nevertheless, throughout the country there are many ruins of ancient towns and villages; it was evidently at one time heavily populated, and so must have been a well-watered region. There are both Arab and French records to show that up to the middle and towards the end of the 18th Century . . . these towns were inhabited by an active farming and trading people; the area, however, became deforested and it has only taken some 200 years to depopulate a country as large as the Union of South Africa . . . First came the shifting cultivator with his axe and fire; secondly, the grazier with his camels, cattle, sheep, and goats . . . and now comes the desert. . .

If such changes take place on the Plains—if we fail to stabilize the dunes in time and allow them to coalesce and cover townships, counties, or groups of counties—no exceptional vision is required to perceive the land anarchy which will follow the probable eastward march of sand across extensive areas of presently productive farms. No one can predict, of course, just how far the dunes may advance with the prevailing winds before nature can check them with vegetation. We do know that in the geologic past sand dunes spread almost across Nebraska before bluestem and other plants finally anchored them.

For a number of years now, the Soil Conservation Service has been studying this problem of shifting dunes and experimenting with a wide variety of control methods. Perhaps the outstanding achievement along this line is the work that has been accomplished near Dalhart, Texas. On one field in this area in 1936, there were 57 newly formed dunes ranging from 1 to 9 feet in height and averaging 161 feet in length and 113 feet in width. Men assigned to the task of stabilizing the dunes worked on the simple theory that winds which pile the soil up in drifts might be used to redistribute it evenly over the area. In November, 1936, the entire area was listed across the direction of the prevailing winds. By March of the following year, it was estimated that approximately 60 per cent of the soil material had been spread back over the field—the intervening or interdune areas—and the dunes decreased in number

³ Ainslie, J. R. 1935 Soil Erosion in Nigeria. British Empire Forestry Conf., South Africa, 1935. Kaduna, Nigeria.

from 57 to 29. In June the whole field was planted to Sudan grass, kafir, black amber sorgo, millet, and broom corn, and further efforts were made to tear down and distribute the remaining high portions of the dunes. Today there is no sign of these dunes whatsoever; even the men who worked on the job are able to point out their former locations only by means of boundary lines indicated by stakes.

More recently the Service has tackled another job of dune stabilization in Bent County, Colorado. Construction of the Caddoa Reservoir in this county by the army engineers involved the relocation of several miles of main track on the Santa Fe Railway. Two routes were available: one required two bridges across the Arkansas River and was through a valuable irrigated section of the valley, and the other was through a dry, sandy area covered by several active dunes ranging from one-half acre to 100 acres in size and from 5 to 20 feet in height. The railroad finally agreed to relocate its tracks through the dune area, at an immense saving to the government, provided the government could succeed in bringing the dunes under control.

In this job, the army engineers called on the Soil Conservation Service for help, and field work was started in October, 1939. Treatment on the active dunes has involved three operations: spreading a thick straw mulch over the entire area; planting the dunes to species of grass that grow well on sandy soil; and packing the mulch down with a heavy roller to cover the grass seed and anchor the mulch by forcing it into the sand. It is still early to make any final predictions; but already grass seedlings planted in March of this year have made considerable growth, and with favorable rainfall, they should become well enough established before winter to anchor the dunes with some degree of permanence.

MEETING INDIVIDUAL PROBLEMS

In discussing some of the latest soil and water conservation developments in this region, only the outstanding types of land treatment have been mentioned. Actually dozens of practices that need not be described in detail are being used over the Plains. If one feature stands out above all others, it is the overwhelming need for a *flexible approach*. Past experience has shown time and again that there is no easy remedy, no panacea, for the land problems of the Plains or any other region. Methods that work wonders on the sandy soils often prove completely inadequate on the "hard" lands (soil of good clay content)—and *vice versa*. The only solution that holds promise of lasting success is to analyze each farm or ranch, both as a variable piece of land and as a business enterprise, and to treat each parcel of land in accordance with its needs and adaptabilities. This principle is and always has been fundamental in the whole Soil Conservation Service program.

The Smoky Hill erosion control and water conservation project in Cheyenne County, Colorado, is especially illustrative of what has been done and what is being done to save the soils of the Plains. Five years ago, this 160,000-acre

area of intermingled farm and range land was rather severely depleted by both wind and water erosion. In many places, more than 6 inches of the surface soil had been scoured away, and numerous hummocks and sand dunes presented a serious obstacle to farming operations. A large percentage of the operators had been forced to abandon their farms and ranches.

When the project started, the most important task was to store up enough water in the soil to provide a cover of protective vegetation. This was accomplished, in the main, by contour furrowing and terracing of range land and contour listing and terracing of farm land. For the last few years, these measures have held virtually all the rainfall and have distributed it so well that green cover is now established over most of the 90,000 acres where conservation measures have been installed. On all these farms and ranches, soil blowing and soil washing have been sharply reduced; and on many the erosion problem has been virtually eliminated.

ECONOMIC ASPECTS

Work of this kind throughout the Plains has produced important benefits over and above physical stability of the land. Examples of increased yields and better incomes already have been cited for the region as the result of soil and water conservation operations. As a natural corollary to this, large numbers of conservation farmers are now operating on a more stable economic basis than ever before. Many who formerly devoted almost all their lands and all their energies to the production of undependable cash crops like wheat are now supplying more of their home needs from the products of their own acres. They are developing gardens, raising poultry, increasing their livestock herds, and bringing idle lands into productive use. The net result of erosion control and water conservation work on farm after farm has been a better diversified, more self-sufficient, and more generally profitable type of agriculture.

The problem of farm abandonment has been especially critical in this region in recent years—perhaps more critical than anywhere else in the country. But surveys show that soil defense work has already helped to bring greater security for people on the land. In ten typical counties of the Southern Great Plains, the Soil Conservation Service recently found that about one farm out of every six has been abandoned in the last few years. Where the land had been treated for soil and water conservation in these same counties, however, the ratio of farm abandonment was less than one in thirty. And in seven of the ten counties surveyed, there was no abandonment whatever on the conservation-treated farms.

Despite all these encouraging results, however, there is no denying the fact that soil and water conservation work, in itself, does not constitute a complete answer to the land troubles of the Plains. In many parts of this region, the problem of land reform is complicated by burdensome taxes, heavy debts, and ill-adapted patterns of ownership. Many farmers and ranchers, for example, simply do not have enough land to adopt a sound program of management no matter how urgently they may desire to protect their fields and ranges.

Operating on cramped, inadequate holdings, they are virtually compelled to overtill or overgraze the land in order to make a decent living.

But progress is also being made toward a solution of this particular problem. In 16 land utilization project areas of the Southern Great Plains, the federal government since 1934 has bought nearly one million acres of land that is unsuitable for cultivation and has developed it as range or community pasture. In these areas, which are now under supervision of the Soil Conservation Service, the newly developed grazing lands are made available to the farmers and ranchers who remain in the area after the restoration work is completed. Thus, men on small farms or ranches are being given a long-needed chance to expand and adopt a more conservative type of land use.

Through the acquisition of land by the government under the land utilization program and with the financial assistance extended to farmers and stockmen by the Farm Security Administration, real progress has been made in certain areas in helping operators to reorganize their small cash crop farms and to increase the size of their operating units. In the reorganized units, the major source of income is from forage crops and livestock rather than from cash grain crops. As a result of such reorganization, land primarily unsuitable for cultivation is restored to grazing use, conservation practices are followed in the use of all lands and in the production of feed crops, and the operators are provided a more stable basis for making a living from the land.

Another government program aimed at relieving some of the pressures that make for land abuse in this region is being carried out under authority of the Pope-Jones Water Facilities Act of 1937. Under provisions of this Act, farmers in 19 areas of the Southern Plains are receiving technical help from the Soil Conservation Service in establishing complete soil and water conservation programs on their land, and loans by the Farm Security Administration are being made for development of water facilities to serve the farm needs. For these people, carefully planned water facilities have helped to lighten the whole routine of living on the land and to place it on a more substantial basis.

But perhaps the most promising development of all in the last few years has been the rapid formation of soil conservation districts both in this region and over the country as a whole. These local subdivisions of government are, in a certain sense, farmer cooperatives established for the purpose of controlling erosion and conserving water resources. All of them embody the same spirit of community enterprise that has been so vividly demonstrated by the district in the western part of Baca County, referred to above. This spirit will be found in the districts of Georgia, Arkansas, Minnesota, and many other states. It is for this reason above all others that confidence is aroused in these districts as a means of attaining our local, state, and national conservation objectives. Probably we have in the state soil conservation district the most promising mechanism that has ever been proposed for introducing soil and water conservation measures rapidly and permanently across the millions of acres in this country now needing treatment.

So far 35 districts have been created in the Southern Plains region covering

several millions of acres. Dozens more are in the process of formation. As rapidly as possible, the Soil Conservation Service is extending assistance to these districts, helping the farmers to plan and establish complete farm programs for soil and water conservation. At the same time, other agencies such as the State Extension Service, the Farm Security Administration, and the Agricultural Adjustment Administration are also making valuable contributions. The whole picture in the districts is one of cooperation—among government agencies as well as among the farmers themselves.

A WORD OF WARNING

If these districts continue to move ahead during the next ten years at the rate that has prevailed since 1937, it is distinctly possible that by 1950, or before, the forces of conservation in the Plains will gain the upper hand over the forces of land depletion. Right now, however, some developments appear to be slowly beginning which might conceivably threaten the whole future of the conservation movement and even wipe out the hard-won gains of the past several years. This spring a large acreage of native grassland that has never been plowed before in the southern part of this region was broken and planted to crops. In itself, this development is serious enough, but the really dangerous aspect of the situation is that it may possibly indicate a trend.

Twenty-five years ago, the farmers of the Great Plains made one of the most costly mistakes in American history. Under the impetus of heavy demands for food products, of war-time prices, and even of government encouragement, they pushed their plows and drills across perhaps 30 million acres that had previously been in native sod. The final result has been written on the face of the land in the widespread dust storms and the intense human suffering of the past several years. That great plow-up contributed largely to this outstanding example of incorrect land use.

Today nations of the Old World are once again at war. Events are moving so fast, taking so many unexpected turns, that nobody can predict the future. If prices should rise sharply and if farmers in the Plains should succumb to the same temptations as they did 25 years ago on anything like a comparable scale, however, there would be every reason to expect a tragic recurrence of a previous tragic land disaster.

Regardless of the outcome of the war, we simply cannot afford to jeopardize our most basic resource. If the need for expanded farm production should arise, expansion *can* be achieved without increasing erosion hazards. But the expansion will have to be planned carefully and not brought about in the pell-mell fashion that prevailed during the last World War. If we plow up lands wholesale with no thought of their inherent capabilities or their susceptibilities to erosion, we shall all eventually become in real truth a poorer nation of people. Whatever the future, we must include in our national policy defense of our land against the forces of exploitation and waste.

EFFECT OF FIRE-HEATING ON THE PROPERTIES OF BLACK COTTON SOIL IN COMPARISON WITH THOSE OF GRAY AND OF HUMUS-TREATED SOILS

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It is well-known that heavy soils such as the black cotton soil of Malwa² very easily lose their tilth on wetting. They become compact and sticky after continuous rains or on irrigation, and later dry to hard clods. Under such conditions, germination of seed is difficult and uncertain, growth is stunted, and yield is reduced. Various diseases also appear, causing much loss. It is, therefore, impossible to grow, at any rate economically, many field, garden, or fruit crops.

During an investigation to control this loss of tilth under field conditions, it was noticed that after being lightly fired, the soil ceases to become sticky when moistened with water. Soil so prepared was then tested for its influence on rain-grown and irrigated crops, both annual and perennial. Without a single exception, improvement in the vigor of growth and fruiting was observed. For instance, in 1933, small plot experiments with cotton gave the results shown in table 1.

Application of fired soil produces a residual effect lasting for at least two years. Fired soil was subsequently used in the Sudan, and yield increases of 25 per cent in seed cotton were obtained (2). Further field trials on a large scale carried out at this Institute have also yielded promising results.³

The soil layers of similar conductivities in the 1933 plots were examined throughout their depth after the cotton harvest in January, 1934, that is, about 7 months after application. The results are given in table 2.

The treatments appear to have markedly influenced the whole soil profile. It was therefore thought necessary to study the changes produced in local soil when subjected to the firing process and to discover whether some of these changes can be held responsible for the differences observed in plant growth. It was expected that such an investigation would reveal the fundamental basis

¹ This work was undertaken at the instance of Y. D. Wad, to whom the authors' grateful thanks are due.

² Malwa is a high level region of nearly 8,000 square miles forming the greater part of the western section of Central India. The plateau is covered principally by black cotton soil, locally known as *mar* or *kali*. The black soil is formed by the disintegration of the Deccan trap prevailing in this region.

³ *Inst. Plant Indus., Indore, Ann. Rpts. 1939-40.*

on which the control of soil tilth depends and might lead to some alternative capable of achieving the same result.

Two other instances of soil, natural and treated, which produced differences in crop growth similar to those produced by the firing process suggested themselves for comparative study:

1. Certain gray-colored soils found in low-lying areas in Malwa are often extraordinarily productive compared to the adjoining black soils from which they are formed. Some of the results obtained by controlled growth of rain-fed cotton in small lysimeter plots (about 9 feet x 7 feet) filled with profile blocks (4 to 6 feet deep) of black and gray soils taken from the field are given in table 3.

2. The permanent manurial small plots (7 feet x 4½ feet) at the Institute, where the soil was mixed throughout the profile down to the *murum* (18 to 24 inches deep) with humic manures in 1933 and was subsequently given annual surface dressings of these manures at 10 tons per acre, have consistently

TABLE 1
Cotton yields on fired and unfired soils, Indore, 1933
Calculated yield of seed cotton in grams per 100 sq. ft.

COTTON VARIETY	UNMANURED			MANURED*		
	Untreated	Heated soil		Untreated	Heated soil	
		6-inch surface layer	50 per cent in 6-inch surface layer		6-inch surface layer	50 per cent in 6-inch surface layer
Indore 1.....	221	456	608	277	762	955
Malvi 9.....	658	1,164	801	910	1,399	1,174

* Farmyard manure applied at the rate of 7.4 tons per acre.

given increased yields due to application of organic manures. A few of the yield data for cotton with the untreated and farm compost treated plots are given in table 4.

In view of the similarity of the foregoing differences in yield, it was considered likely that some common features influencing crop growth might be found in the fired soil, the gray soil, and the humus-treated soil. A comparison with oven-heated soils also seemed desirable to gain a better understanding of the changes in soil properties as a result of firing.

LITERATURE

In parts of India, it has long been the practice to grow rice in seedbeds prepared by burning waste organic matter on the ground. This process, called *rab*, is considered by the cultivator to be of the greatest importance, and the collection of the necessary materials is, in fact, his chief occupation from December to May. The effect of such firing on the crop has always been

markedly beneficial. The phenomenon appears to be common throughout the tropical and subtropical regions of the world. Hill tribes bring forest

TABLE 2
Soil profile changes produced by cropping and treatments

HEATED SOIL							UNTREATED CONTROL						
Depth	Hygroscopic moisture	Nitrogen	Carbon	Carbon-nitrogen ratio	Conductivity ratio 8 days 24 hours	Base-exchange capacity	Depth	Hygroscopic moisture	Nitrogen	Carbon	Carbon-nitrogen ratio	Conductivity ratio 8 days 24 hours	Base-exchange capacity
inches	per cent	per cent	per cent			m.e./100 gm.	inches	per cent	per cent	per cent			m.e./100 gm.
<i>Variety—Indore 1</i>													
Unmanured													
0-6	3.46	0.14	0.22	1.8	2.2	66.2	0-3	5.16	0.10	0.26	2.6	1.6	82.2
6-9	4.56	0.09	0.25	2.8	2.1	74.1	3-6	4.33	0.06	0.26	4.3	1.9
9-15	5.32	0.09	0.13	1.4	1.8	72.9	6-9	4.90	0.07	0.40	5.7	2.0	86.3
15-21	4.86	0.08	0.25	3.1	1.8	88.9	9-15	6.06	0.09	0.30	3.3	2.2	81.1
21-24	5.01	0.07	0.25	3.6	1.8	74.3	15-18	5.80	0.09	0.24	2.7	1.9	74.9
.....	18-24	5.58	0.06	0.34	5.7	2.0	74.7
Manured													
0-3	4.33	0.09	0.31	3.5	2.1	67.8	0-3	4.20	0.10	0.31	3.1	2.7	66.9
3-15	4.51	0.08	0.25	3.1	2.1	61.6	3-12	5.15	0.08	0.14	1.8	2.6	69.7
15-21	3.84	0.06	0.25	4.2	1.8	66.3	12-24	4.60	0.08	0.15	1.9	2.2	67.8
21-24	4.78	0.06	0.29	4.8	1.9	63.7
<i>Variety—Malvi 9</i>													
Unmanured													
0-3	5.28	0.09	0.57	6.3	2.2	64.9	0-3	5.61	0.14	0.59	4.2	1.8	69.4
3-15	7.29	0.06	0.46	7.7	2.4	74.5	3-9	5.58	0.11	0.60	5.5	2.4	71.2
15-18	0.08	0.69	8.6	2.0	72.4	9-15	5.96	0.10	0.37	3.7	2.2	68.5
18-24	7.88	0.07	0.39	5.6	2.1	78.5	15-18	5.84	0.10	0.34	3.4	2.7	73.6
.....	18-21	6.04	0.10	0.45	4.5	1.9	74.1
.....	21-24	6.14	0.11	0.42	3.8	1.9	74.6
Manured													
0-3	5.52	0.10	0.73	7.3	2.2	73.0	0-3	6.49	0.14	1.07	7.6	2.5	71.6
3-9	7.69	0.09	0.77	8.6	2.3	69.4	3-9	7.22	0.11	0.74	6.7	2.6	72.2
9-15	7.92	0.07	0.38	5.4	2.1	67.2	9-15	7.70	0.08	0.69	8.6	2.3	72.2
15-21	6.29	0.07	0.33	4.7	1.9	70.6	15-24	7.31	0.07	0.59	8.4	2.1	71.3
21-24	7.42	0.07	0.53	7.6	1.5	80.0

land or scrub into cultivation by a process of firing the ground vegetation. Graziers set fire to grassland occasionally; the grass that grows in the following year is always sweeter and gives better feed for the cattle.

In temperate countries, the old method of "paring and burning" heavy soil when pasture is being converted into arable land or when it is desired to improve heavy clay may perhaps be considered similar to the *rab* process. In this case, clay is burned in heaps at a low temperature and then spread on the land. As a result, the soil is sweetened and cleaned, parasites and weeds are killed, the mechanical texture is improved, and the available mineral plant foods are increased (32).

Heating the soil has also been frequently recommended as a method to be used in garden and greenhouse cultivation (26, 27, 28).

In a scientific enquiry on the causes that make the *rab* system of cultivation efficient, Mann *et al.* (19) have reported more water-soluble solids, both

TABLE 3
Cotton yields on black and gray soils
Calculated yield of seed cotton in grams per 100 sq. ft.

YEAR	COTTON VARIETY	BLACK SOIL WELL-DRAINED SHALLOW FIELD, INDORE	GRAY SOIL TANK AREA, BADNAWAR, DEHAR
1933	Malvi 9	212	1,194
1933	Indore 1	110	1,332
1936	Indore 1	117	1,165
1938	Cwn. 520	25	201

TABLE 4
Cotton yields on humus-treated and untreated soils
Calculated yield of seed cotton in grams per 100 sq. ft.

YEAR	COTTON VARIETY	UNTREATED SOIL	SOIL TREATED WITH FARM COMPOST
1933	Malvi 9	1,170	2,167
1934	Malvi 9	656	1,396
1934	Indore 1	165	749
1936	Indore 1	449	1,305

organic and inorganic; greater permeability; less viscid matter; and decreased oxygen-absorption capacity of the heated soils. These authors have also observed that adding safflower cake or gypsum produces a flocculating effect similar to heating. Klein (16) and, later, Lebedjantzev (17) have shown that the drying of a soil previous to planting has a beneficial effect on plant growth; bacterial activity and nitrification increase, reaching a maximum with three dryings. Steenkamp (31) has found that when soil is dried, there is a distinct cementation of fine material as well as a flocculating effect which, under field conditions, renders the soil easy to work. With pasture soils the degree of saturation decreases on dehydration but increases with cultivated soils. On the other hand, Achromeiko (1) asserts that the absorptive capacity for

bases does not decrease on drying, and attributes the decrease observed by other workers to a change in the aggregation of the finer soil particles and a failure under these conditions thoroughly to remove the bases on account of the more rapid percolation of the extractants through the dried samples. Barnette and co-workers (4, 12) have observed that forest soils subject to frequent fires are less acid and have a higher percentage of replaceable calcium, total nitrogen, and organic matter and that the chemical changes due to firing are limited to the upper 4 to 6 inches of the soil. In a study of the effect of annual grass fires on the organic matter and other constituents of virgin long-leaf pine soils, Greene (10) also found that burned-over soil is richer in organic matter than soils protected from fire. The quantity of forage growth on the ungrazed burned areas is more than double that on the unburned areas. Fowells and Stephenson (9) have shown that though burning partly destroys the organic matter, the ash constituents of burnt soils stimulate nitrification and add to the mineral nutrients in the soil. From impact measurements, it has been reported by Andrews and Clouston (2) that the firing of the top few inches of the soil seems to have some effect on the resistance to penetration of the subsoil.

The foregoing observations suggest that the increased fertility of heated soil may be due to an increase in available mineral and organic plant foods, a change in the physical texture, and, possibly also, changes in the micro-organisms of the soil. This paper attempts to throw further light on the subject with special reference to the similarity or dissimilarity of the changes brought about by continuous heavy manuring and between gray and black soils.

EXPERIMENTAL

Materials

Group 1—Heated and fired soils. Loose black soil from the surface of a field of medium fertility was collected and fired according to the following procedure:

Stiff and thin stalks of crops such as those of *tur* (*Cajanus indicus*), sann hemp or cotton, reeds, and twiggy jungle plants such as *baer* (*Zizyphus jujuba*)⁴ are arranged to form a circular layer 8 to 10 feet in diameter and about 5 inches thick, the butt ends of the stalks being kept outward in the form of a cartwheel. A bundle of fuel about a foot in diameter is placed vertically in the center, and around it, but 3 to 4 inches away from it, is placed a layer of soil about 3 inches thick in the form of a ring, at least 6 inches of fuel being left uncovered all around the heap. On top of this ring of soil another layer of fuel is placed in the same manner as the first layer. Upon this is laid another ring of soil around the upright central bundle of fuel, which is kept projecting above the heap by means of fresh additions of fuel if necessary and which thus acts as a chimney. A third layer of fuel followed by a third ring of soil with a fourth cover of fuel completes the heap. Each successive ring and layer becomes slightly smaller in diameter so that the heap tapers upward into a cone.

The heap is lighted at the base, starting at the side away from the wind and thence on both

⁴ Softer wastes tend to give way under the weight of the soil and therefore fail to burn because of lack of air.

sides toward the wind. The first fierce flames die down in about 15 minutes, after which the burning is slow and smoldering. This is allowed to continue for about 24 hours, when the heap is broken.

About 45 pounds of fuel is required to heat soil weighing about 400 pounds. Examination of soil from a number of heaps burned in this way has shown that a properly burned heap contains 15 per cent of over-burned (brick colored), 23 per cent of under-burned (unchanged in color), and 62 per cent of properly burned (light or dark bluish purple in color) soil.

Properly fired soil breaks into granular powder and, unlike the original black soil, when put in water, does not become sticky but gives a suspension with purplish red flakes which do not make the water permanently turbid. Another reliable test of a properly fired soil is to add a drop or two of water to a lump, when it will be seen to crumble into a fine powder. A heap of the dimensions given yields about one-half ton of correctly fired soil and can be successfully managed by one male and two female laborers working for 3 to 4 hours. The collection of fuel and soil takes nearly an equal amount of time. This quantity will be sufficient for application to an area of more than 5 acres. It is extremely important that the proportion of over-burned soil, as indicated by its brick-red color, should be kept within low limits. Over-burned soil, when mixed with raw black cotton soil, packs and cakes even more than does black soil alone. Under such conditions, crops naturally suffer very severely.

The properly fired portions of the soil were separated, freed from ash contamination as far as possible, and, after being crushed, were passed through a round 1-mm. mesh sieve. Portions of the untreated soil were also passed through the sieve and oven-heated for 24 hours at (a) 90°–100°C., and (b) 140°–150°C. There were thus four soils in this group; namely, untreated, lightly heated, strongly heated, and fire-heated.

Group 2—Soils untreated and treated with compost. The loose surface soils from the control plots and the farm compost-treated plots of the permanent manurial beds in the Institute were collected, air-dried, and graded through a 1-mm. sieve.

Group 3—Black and gray soils. Samples were collected as in the previous group from the lysimeter plots maintained at the Institute.

Methods

The following determinations were made according to methods individually cited:

Moisture [A.O.A.C. (3)].

Hygroscopic moisture and moisture at sticky point [Keen and Coutts (13)].

Keen-Raczkowski measurements; namely, moisture content of saturated soil, apparent density, pore space, specific gravity, and swelling capacity (7, 14).

Conductivity by Wheatstone bridge (30).

pH by glass electrode using an electrometer valve pH meter.

Mechanical analysis [Robinson (23)].

Loss on ignition [Wright (33)].

Carbon, nitrogen, and carbon-nitrogen ratio [Robinson *et al.* (24)].

Available potash and phosphoric acid by Dyer's method (8) followed by estimation of potassium by the cobaltinitrite method (18) and of phosphoric acid by Pemberton's method (3).

Total exchange capacity [Puri (20)].

Replaceable sodium, potassium, and magnesium [Puri (21)] and replaceable calcium [Puri (22)].

Wilting coefficient was calculated from the hygroscopic moisture by use of the factor 1.47 (6). Dispersion coefficient was obtained from the formula $\frac{D-S}{D} \times 100$, where D is percentage of clay (0.002 to 0 mm.) obtained by ultimate analysis and S is the percentage of clay obtained in aggregate analysis (29). The values for D and S were determined by Bouyoucos' hydrometer method (5) in the completely dispersed and water-dispersed samples respectively. Index of texture was calculated according to Hardy (11).

TABLE 5
Hygroscopic moisture, moisture at sticky point, and "box constants"

DESCRIPTION	HYGROSCOPIC MOISTURE	WILTING COEF- FICIENT	MOISTURE AT STICKY POINT	WEIGHT OF 100 CC. OF SOIL	MOISTURE CON- TENT OF SAT- URATED SOIL	PORE SPACE PER 100 CC.†	SPECIFIC GRAVITY†	VOLUME EX- PANSION PER 100 CC.
	per cent*	per cent*	per cent*	gm.†	per cent†	cc.		cc.†
<i>Group 1</i>								
Untreated control.....	9.51	13.99	42.35	135.2	64.30	59.78	1.487	39.67
Lightly heated, (90-100°C.) ..	7.96	11.76	44.28	124.9	72.72	60.03	1.502	46.96
Strongly heated, (140-150°C.).	7.81	11.48	44.07	124.5	80.10	60.03	1.648	50.44
Fire-heated.....	5.45	8.01	32.60	136.7	57.61	57.59	1.359	38.25
<i>Group 2</i>								
Untreated control.....	8.45	12.42	41.28	136.7	58.36	56.15	1.360	38.67
Treated with compost.....	7.75	11.39	41.87	133.6	63.64	56.23	1.428	42.58
<i>Group 3</i>								
Black soil.....	8.76	12.88	43.62	133.9	65.21	58.38	1.403	39.13
Gray soil.....	4.98	7.37	31.73	133.9	57.14	57.48	1.352	33.82

* On oven-dry basis.

† On air-dry basis.

RESULTS

For convenience of presentation, the results, which represent averages of duplicate determinations, have been grouped together in tables 5 to 8 and are discussed here in that order.

Hygroscopic moisture and "box constants"

There is throughout a remarkable similarity in the differences, shown in table 5, between black and gray soils (group 3) and those brought about by fire-heating (group 1). Thus, the reduction is appreciable in both cases in the values for hygroscopic moisture and moisture at sticky point, and the fall in the values for maximum water-holding capacity, pore space, specific gravity,

and volume expansion is slight though definite. The change in apparent density is virtually nil in both cases.

The changes produced as a result of humus application (group 2) are similar to those of fire-heating in the case of hygroscopic moisture only, a slight increase or no change occurring in the other properties.

The hygroscopic-moisture values for the oven-heated soils (group 1) are intermediate between that for the untreated soil and that for the fire-heated soils. In all other cases, oven-heating brings about an increase over the control values.

TABLE 6
Mechanical analysis, index of texture, and dispersion coefficient

DESCRIPTION	MOISTURE	LOSS IN SOLUTION	COARSE SAND	FINE SAND	SILT	FINE SILT	CLAY	INDEX OF TEXTURE	PORE SPACE CLAY	DISPERSION CO-EFFICIENT		
										D	S	$\frac{D-S}{D} \times 100$
<i>Group 1</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>					
Untreated control .	7.28	2.49	1.25	7.04	13.51	13.84	48.33	40.56	1.24	47.5	22.6	52.4
Lightly heated . . .	6.69	3.93	1.07	9.74	22.06	23.26	19.67	41.96	3.05	20.1	9.0	55.2
Strongly heated . . .	6.49	4.64	0.75	9.78	21.53	23.09	21.93	41.82	2.74	22.4	8.4	62.5
Fire-heated	4.52	2.33	5.40	11.30	18.80	22.83	29.45	29.10	1.96	30.1	5.1	83.1
<i>Group 2</i>												
Untreated control .	6.54	2.57	1.55	8.80	16.63	15.77	38.85	39.07	1.95	40.4	19.5	52.0
Treated with com- post	6.37	2.93	0.86	9.03	11.24	24.45	36.77	39.76	1.53	36.5	13.1	64.1
<i>Group 3</i>												
Black soil	6.79	2.21	3.18	10.33	17.06	19.46	33.15	40.72	1.76	35.7	19.2	46.2
Gray soil	4.16	1.84	2.38	23.39	19.73	20.61	20.52	26.35	2.80	23.2	8.4	72.4

It would appear, on the whole, that oven-heating results in a slight swelling of the soil and that direct firing shrinks the soil somewhat (*cf.* weight of, and maximum water absorption by, unit volume of soil, pore space, and shrinkage). Effects similar to oven-heating are probably also produced in the humus-treated soil.

Mechanical analysis, index of texture, and dispersion coefficient

The results of the mechanical analysis (table 6) show that there is a marked decrease in the clay content and a corresponding increase in the contents of silt, fine silt, and fine sand as a result of heating (group 1). The magnitude of the difference is less with the fire-heated soil as compared with the oven-heated specimens except for the fine sand fraction, where the increase is greater

in the fire-heated soil. This point is of some significance, since fine sand does not swell or shrink as a result of moisture changes and yet holds moisture very well.

A similar decrease in clay content and increases in other fractions are observed with the gray soil as compared to the black soil (group 3); among the coarse fractions, the difference is again pronounced in the fine sand fraction.

In the compost-treated soil, however, the decrease in clay content is small (group 2) and is accompanied by an increase only in the fine silt and fine sand fractions, the other two fractions decreasing.

The figures for texture index (11) reveal that though no appreciable difference is brought about by oven-heating or by humus application, fire-heating lightens the soil considerably. The gray soil is nearly equally light-textured.

The ratio of pore-space to clay is increased by all the treatments in group 1, indicating that the structure is improved: the increase is greater in the oven-heated samples than in the fired soil. The dispersion coefficients, however, show that the extent of aggregation increases steadily with lightly heated, strongly heated, and fire-heated soils. Structure improvement is also pronounced in the humus-treated and in the gray soils.

Exchange capacity, replaceable bases, pH, and conductivity

The exchange capacity of the soil (table 7) is lowered as a result of heating, light heating being more effective than strong heating. Fire-heating has the greatest lowering effect. Compared to the black soil, gray soil has also a low exchange capacity. Compost treatment likewise lowers the exchange capacity of the soil. This is contrary to the generally held view that humus increases the capacity of the soil to hold exchangeable bases. It is possible, however, to obtain such an effect on the surface layers because the crumb structure developed might have allowed the finer clay to move rapidly into the lower layers, thus reducing the exchange capacity of the surface soils. In fact, there is a close parallelism in all the groups between the exchange capacity, the clay content (table 6), and the hygroscopic moisture (table 5).

Oven-heating has progressively increased all the individual bases except magnesium, which decreased. Firing has reduced calcium and magnesium but increased sodium and potassium. The results are similar with the gray soil, the difference in calcium being considerable in this case. Compost treatment has increased all bases except magnesium.

Percentage base saturation is over 100 for all black soils but is slightly lower for the gray soil. Firing also lowers base saturation to some extent. The ratio of calcium to magnesium is increased and that of calcium to sodium decreased as a result of firing or of humus treatment and in the gray soil.

The pH is increased by heating, the increase being greatest with the fire-heated soil. This may be due in part to contamination with plant ash during firing, but the change in reaction is parallel to the changes in exchangeable sodium and potassium contents. The pH is lower in compost-treated and in gray soils.

TABLE 7
Exchange capacity, replaceable bases, pH, and conductivity

DESCRIPTION	EXCHANGE CAPACITY $m.e./100$ gm.	REPLACE- ABLE SODIUM $m.e./100$ gm.	REPLACE- ABLE POTASSIUM $m.e./100$ gm.	REPLACE- ABLE MAGNE- SIUM $m.e./100$ gm.	REPLACE- ABLE CALCIUM $m.e./100$ gm.	REPLACEABLE CALCIUM MAGNESIUM	REPLACEABLE CALCIUM SODIUM	BASE SATURA- TION per cent	pH	CONDUCTIVITY			
										After 24 hours $mhos$ $\times 10^{-4}$	After 8 days $mhos$ $\times 10^{-4}$	"7-day in- crease" $mhos$ $\times 10^{-4}$	Ratio 8 days 24 hours
<i>Group 1</i>													
Untreated control.....	40.8	0.25	0.10	3.83	37.5	9.8	149.9	102.0	8.17	139	233	94	1.68
Lightly heated.....	38.8	0.25	0.17	3.67	38.6	10.6	153.2	109.9	8.43	250	313	63	1.25
Strongly heated.....	39.6	0.29	0.19	3.76	42.7	11.4	151.6	118.6	8.53	385	385	..	1.00
Fire-heated.....	37.4	0.32	0.28	2.67	32.9	12.3	102.2	96.6	8.73	400	400	..	1.00
<i>Group 2</i>													
Untreated control.....	38.5	0.19	0.24	3.21	35.4	11.0	185.0	101.2	9.11	116	200	84	1.72
Treated with compost.....	33.9	0.21	0.25	2.99	45.5	12.5	176.0	120.1	8.79	139	238	99	1.71
<i>Group 3</i>													
Black soil.....	35.0	0.23	0.36	3.89	33.3	8.6	144.5	108.0	9.02	217	323	106	1.49
Gray soil.....	27.5	0.27	0.40	2.18	23.5	10.3	87.0	96.0	8.96	144	238	94	1.65

Conductivity is increased by heating, stronger heating being more effective. Fire-heating gives the maximum conductivity. Compost treatment also increases conductivity, though not appreciably. Gray soil has a lower conductivity than its control. Strongly heated and fired soils reach their maximum conductivity within 24 hours, thus suggesting the release of salts during heating.

Carbon, nitrogen, and available potassium and phosphorus

There is no change in the carbon content as a result of light or strong heating but there is a slight loss of nitrogen, the loss being greater with stronger heating. As a result of fire-heating, both carbon and nitrogen are lost in appreciable quantities, and the carbon-nitrogen ratio is lowered. The increase in organic matter content of burned-over forest soils reported by other workers (4, 10) may be due to enrichment by partly burned materials.

Treatment with compost continuously for several years has increased the carbon content of the soil nearly threefold, while nitrogen is increased by about 15 per cent. The ratio of carbon to nitrogen is higher also in the gray soil, though the nitrogen content is the same as in the black soil.

Gray soil has the highest available potash and phosphoric acid. The black soils or their modifications show only minor variations in available phosphorus or potassium, a slight increase in the former and a decrease in the latter occurring as a result of fire-heating or of compost treatment. That black soils have the maximum content of available potash is possible, inasmuch as composting does not increase it. Although firing, by itself, has not increased the available potassium or phosphorus of the soil, it must be pointed out that added potassic and phosphatic fertilizers will be made more available by this treatment because of lowered adsorptive capacity.

GENERAL DISCUSSION

The main effects of fire-heating of black cotton soil on crop growth appear to be due to an improvement in the physical properties of the soil. The colloidalilty of the soil is greatly reduced by firing, and there is a corresponding increase in the coarse fractions. This is doubtless partly due to the formation of stable aggregates, which promote better aeration, improve texture, and increase permeability. There is also an indication⁵ that fired soil can be successfully used to control the rise of subsoil water.

Treatment with compost also results in improvement of structure and other desirable physical properties, but the better crop yield obtained on application of humic manures is also partly due to the supply of organic plant food (table 8).

The remarkable similarity in the properties of fired and gray soils can perhaps be best understood when it is realized that these gray soils are only transformations of black soil formed either in low-lying areas adjoining black

⁵ Unpublished data.

soil or after continuous heavy manuring and irrigation. The exact nature of these changes is not well understood and is in course of study at this Institute.

It may be mentioned here that increased yields of cotton have also been obtained in field trials with surface applications of gray soil as with fire-treated soil. Fired or gray soil can be applied economically in a variety of ways to improve the yielding capacity of black cotton soils. Thus, it can be filled in trenches alongside alternate rows of cotton, at the bottom of furrows, in ridges, and in bores made by an earth auger.

The changes that accompany the drying or heating of soils are very complex. As pointed out by Kelley and McGeorge (15), among the many factors involved are flocculation of colloids, oxidation, deoxidation, decomposition,

TABLE 8
Carbon, nitrogen, and available potassium and phosphorus

DESCRIPTION	CARBON	NITROGEN	CARBON-NITROGEN RATIO	AVAILABLE K_2O	AVAILABLE P_2O_5
	<i>per cent*</i>	<i>per cent*</i>		<i>per cent*</i>	<i>per cent*</i>
<i>Group 1</i>					
Untreated control.....	0.64	0.14	4.57	0.0034	0.013
Lightly heated.....	0.68	0.13	5.23	0.0036	0.012
Strongly heated.....	0.64	0.12	5.33	0.0034	0.013
Fire-heated.....	0.36	0.09	4.00	0.0033	0.015
<i>Group 2</i>					
Untreated control.....	0.46	0.13	3.54	0.0044	0.012
Treated with compost.....	1.24	0.15	8.27	0.0040	0.015
<i>Group 3</i>					
Black soil.....	0.82	0.13	6.31	0.0035	0.011
Gray soil.....	0.93	0.13	7.15	0.0045	0.108

* On oven-dry basis.

dehydration, and the attendant physical alterations of the soil film. Lebedjantzev (17) has concluded from experimental data that the process of drying is a powerful factor, determining to a large extent the fertility of the soil under natural conditions. It probably plays an important and hitherto unknown role in all the processes of increasing the soil fertility by means of mechanical cultivation of soil.

According to Russell (25), natural heating of the soil can be brought about by proper adjustment of slope and moisture for maximum absorption of the sun's heat. Raw organic matter can also be ploughed in and allowed to decompose *in situ*. The heat of fermentation can be increased by addition of inorganic substances to stimulate microorganic activity.

There is at least one characteristic feature of the tropical black cotton soil which, by a process of natural heating, restores the fertility from time to time. Because of the heavy character of the clays and the extreme variations in

swelling and shrinkage of these soils, wide cracks are formed in the field during the summer fallow. Many of these cracks exceed several inches in breadth and extend several feet downward. During the hot summer months when these cracks develop, a thorough heating and consequent weathering of the subsoil layers occur. These may produce changes in soil properties similar to those which result from firing.

SUMMARY

Light fire-heating of the surface layers of black cotton soils greatly increases crop growth and yield. Similar differences in yield are obtained (a) by surface applications of gray soil, which occurs naturally in many low-lying areas adjoining black cotton tracts, and (b) by heavy application of humic manures throughout the profile of the black cotton soil.

A most suitable technic of fire-heating black cotton soil has been evolved and is described.

A study has been made of the changes in some of the properties of the black soil as a result of lightly heating at 90°–100°C., strongly heating at 140°–150°C., and fire-heating. These changes are compared with the changes attendant on application of humic manures to the soil and with the differences between black and gray soils.

Fire-heating results in a marked improvement in the physical texture and degree of aggregation of the soil colloids and is also followed by a slight decrease in exchange capacity and in replaceable calcium and magnesium, an increase in replaceable sodium and potassium and in total soluble salts, and a loss in organic matter.

Oven-heating somewhat improves the structure of the soil but has no appreciable effect on texture. The changes in replaceable bases or in organic matter are also insignificant.

There is a remarkable similarity in the differences between black and gray soils and in the changes brought about by fire-heating of black soil.

Treatment with humic manure improves structure and other physical properties, but though the beneficial effects on crop growth with fired or gray soil are essentially due to improvements in the physical properties, the effect of humic manure is due in part to organic plant food supply.

REFERENCES

- (1) ACHROMEIKO, A. I. 1931 Influence of drying of soils on their absorptive capacity. *Ztschr. Pflanzenernähr., Düngung, u. Bodenk.* (A) 19: 36–43.
- (2) ANDREWS, F. W., AND CLOUSTON, T. W. 1937 The effect on shoot and root development of "firing" the surface soil. *Sudan Govt. Dept. Agr. and Forests Ann. Rpt.* 1937 (pt. 2): 39.
- (3) Association Official Agricultural Chemists 1935 Official and Tentative Methods of Analysis, ed. 4. Washington, D. C.
- (4) BARNETTE, R. M., AND HESTER, J. B. 1930 Effect of burning upon the accumulation or organic matter in forest soils. *Soil Sci.* 29: 281–284.
- (5) BOUYOUCOS, G. J. 1934 A comparison between the pipette method and the hydrometer method for making mechanical analyses of soil. *Soil Sci.* 38: 335–343.

- (6) BRIGGS, L. J., AND SHANTZ, H. L. 1912 The wilting coefficient for different plants and its indirect determination. U. S. Dept. Agr. Bur. Plant Indus. Bul. 230.
- (7) COUTTS, J. R. H. 1930 "Single value" soil properties: A study of the significance of certain soil constants: III. Note on the technique of the Keen-Rackowski box experiment. *Jour. Agr. Sci.* 20: 407-413.
- (8) DYER, B. 1894 On the analytical determination of probably available "mineral" plant food in soils. *Trans. Chem. Soc.* 65: 115-167.
- (9) FOWELLS, H. A., AND STEPHENSON, R. E. 1934 Effect of burning on forest soils. *Soil Sci.* 38: 175-181.
- (10) GREENE, S. W. 1935 Effect of annual grass fires on organic matter and other constituents of virgin long leaf pine soils. *Jour. Agr. Res.* 50: 809-822.
- (11) HARDY, F. 1928 An index of soil texture. *Jour. Agr. Sci.* 18: 252-256.
- (12) HEYWARD, F., AND BARNETTE, R. M. 1934 Effect of frequent fires on the composition of forest soils in the long leaf pine region (of Florida). Fla. Agr. Exp. Sta. Bul. 265.
- (13) KEEN, B. A., AND COUTTS, J. R. H. 1928 "Single value" soil properties: A study of the significance of certain soil constants. *Jour. Agr. Sci.* 18: 740-765.
- (14) KEEN, B. A., AND RACZKOWSKI, H. 1921 The relation between the clay content and certain physical properties of a soil. *Jour. Agr. Sci.* 11: 441-449.
- (15) KELLEY, W. P., AND MCGEORGE, W. 1913 The effect of heat on Hawaiian soils. Hawaii Agr. Exp. Sta. Bul. 30.
- (16) KLEIN, W. A. 1915 Studies in the drying of soils. *Jour. Amer. Soc. Agron.* 12: 49-77.
- (17) LEBEDJANTZEV, A. N. 1924 Drying of soil as one of the natural factors in maintaining soil fertility. *Soil Sci.* 18: 419-447.
- (18) LEWIS, A. H., AND MARMOV, F. B. 1933 Determination of potassium by the cobaltinitrite method and its application in agricultural analysis. *Jour. Soc. Chem. Indus.* 52: 177T-182T.
- (19) MANN, H. H., JOSHI, N. V., AND KANITKAR, N. V. 1912 The *rab* system of rice cultivation in western India. *India Dept. Agr. Mem., Chem. Ser.* 2 (3): 141-191.
- (20) PURI, A. N. 1934 A new method of estimating base exchange capacity of soils. *Soil Sci.* 37: 105-108.
- (21) PURI, A. N. 1935 A new method of estimating exchangeable bases in soils. *Soil Sci.* 40: 159-163.
- (22) PURI, A. N. 1936 Estimating exchangeable calcium and other cations in soils. *Soil Sci.* 42: 47-59.
- (23) ROBINSON, G. W. 1933 The dispersion of soils in mechanical analysis. Imp. Bur. Soil Sci. Tech. Commun. 26.
- (24) ROBINSON, G. W., McLEAN, W., AND WILLIAMS, R. 1929 The determination of organic carbon in soils. *Jour. Agr. Sci.* 19: 315-324.
- (25) RUSSELL, E. J. 1934 Heating of soil. *Jour. Roy. Agr. Soc. England* 95: 365.
- (26) RUSSELL, E. J. 1934 Soil Conditions and Plant Growth, ed. 6. Longmans Green & Co., London.
- (27) RUSSELL, E. J., AND HUTCHINSON, H. B. 1909 The effect of partial sterilisation of soil on the production of plant food. *Jour. Agri. Sci.* 3: 111-144.
- (28) RUSSELL, E. J., AND HUTCHINSON, H. B. 1913 The effect of partial sterilisation of soil on the production of plant food: II. The limitation of bacterial numbers in normal soils and its consequences. *Jour. Agr. Sci.* 5: 152-221.
- (29) RUSSELL, E. W. 1938 Soil structure. Imp. Bur. Soil Sci. Tech. Commun. 37.
- (30) SEN, A., AND WRIGHT, C. H. 1931 The electrical conductivity of aqueous soil suspensions as a measure of soil fertility. *Jour. Agr. Sci.* 21: 1-13.
- (31) STEENKAMP, J. L. 1928 The effect of dehydration of soils upon their colloid constituents: I, II, III. *Soil Sci.* 25: 163-182; 239-251; 327-332.
- (32) VOELCKER, A. 1857 On paring and burning. *Jour. Roy. Agr. Soc. England* 18: 342-368.
- (33) WRIGHT, C. H. 1939 Soil Analysis, ed. 2. Thomas Murby & Co., London.

CROP PRODUCTION IN ARTIFICIAL CULTURE SOLUTIONS AND IN SOILS WITH SPECIAL REFERENCE TO FACTORS INFLUENCING YIELDS AND ABSORPTION OF INORGANIC NUTRIENTS¹

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The past decade has witnessed the arousal of great popular and scientific interest in artificial media (water culture, sand culture, and gravel culture) as an alternative to soil for crop production. By 1937 the discussion of the commercial possibilities of artificial nutrient media assumed scarcely less than world-wide dimensions and raised several questions of considerable interest to plant physiology and soil science. Among these the following were prominent:

(a) What are the inherent productive capacities of a fertile soil in comparison with those of a favorable nutrient solution?

(b) What are the limiting factors in crop production in a fertile soil, and to what extent can they be controlled in artificial nutrient media?

(c) What is the water economy of plant products grown by these two methods?

(d) To what extent can the dietetic quality of plant products, as reflected in mineral composition and vitamin content, be influenced by the nutrient medium?

The present investigation was undertaken to gain information bearing on these questions and to provide a basis for an independent appraisal of the possibilities of using artificial nutrient media for crop production. This paper deals with the first three questions, primarily from the general point of view of plant nutrition rather than that of the commercial utilization of water culture methods in growing crops. A previous popular publication (9) has presented a discussion of this latter aspect, together with information intended for the use of amateurs. A discussion of the effect of mineral nutrition on the composition of plants is reserved for another publication.

An attempt was also made to arrive at quantitative concepts with respect to the absorption of nutrients by plants at various stages of development and under the influence of increased temperature and oxygen supply around the roots. This problem was approached through periodic analyses of nutrient

¹ A preliminary report including some of the data presented here appeared in *Science* (3).

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³ Contribution from the Divisions of Truck Crops and Plant Nutrition, College of Agriculture.

solutions and by keeping a balance sheet of the quantities of nutrients supplied and absorbed from time to time by the plants.

Since much of the current discussion of the potentialities of the water culture method is based on work with the tomato, this was adopted as the test plant for the investigation.

The results reported here were gathered in the course of two experiments covering approximately one calendar year from August to August. In the first experiment, which will henceforth be referred to as the fall-winter one, the plants were set out in the respective media on August 5 and 6 and were grown until January 31. The second experiment, which will henceforth be known as the spring-summer one, lasted from February 14 to August 23.

EXPERIMENTAL METHODS

The maintenance of a similar climatic environment and of identical spacing and cultural practices for plants grown in soil and in artificial nutrient media was regarded as essential to any valid comparison of yields. To that end the soil and the sand and nutrient solution cultures were placed side by side in a Berkeley (California) greenhouse. The temperature was maintained approximately uniform throughout the greenhouse, but fluctuations of light, determined by the position of each tank relative to others, were unavoidable. The greenhouse extended from north to south, and the tanks were placed in a row, at right angles to the length of the greenhouse, i.e., each tank extended from east to west.

The surface dimensions and the spacing of all tanks as well as the number of plants grown in them were the same as those in the experiment of Gericke and Tavernetti (8). This arrangement was selected in order to provide a comparison of yields from soil and from sand and water cultures under our conditions with the high yields of tomatoes in water culture reported by these investigators on the basis of 25 square feet of water surface.

All tanks used for soil and for sand and nutrient solution cultures had surface dimensions of 10 x 2.5 feet. For nutrient solution cultures the tanks (black iron) were 8 inches deep and held approximately 460 liters of solution. For the soil and the sand, redwood tanks 2 feet deep were used. These were set in excavations in such a manner that at the beginning of the experiments plants in all cultures were at the same vertical level from the ground. The soil was placed directly over the greenhouse excavation in a bottomless redwood tank. For the sand, a redwood tank with a perforated bottom was used, a layer of gravel 2 inches thick being placed underneath it to insure good drainage.

Plants

All plants were started from seed in ordinary river sand, in which they were grown for about 8 weeks and then transplanted into the respective media. The porous bed for supporting the plants placed over the water culture tanks was

similar to that described by Gericke and Tavernetti (8), except that rice hulls were used instead of sawdust. As the plants grew larger they were pruned to a single stem and trained upward, being allowed to extend to the full height of the greenhouse. They were supported by twine attached to the roof at one end and to the tank at the other.

Twenty plants were grown in each tank. In the fall-winter experiment all plants were of the Crackerjack (Earliana) variety; in the spring-summer experiment 10 plants in each tank were of the Crackerjack and 10 of the Lloyd Forcing variety.⁴

The large area of each culture precluded replication of treatments sufficient to permit statistical evaluation of yields. The intent was to determine general trends and differences of large magnitude in crop-producing power of different media.

Nutrient media

The nutrient solution, including the micronutrient supplements (solutions A4 and B7) used in the water and sand culture tanks, was previously described (1). In its preparation distilled water was employed throughout. The proportions of the chief nutrient ions, expressed as milliequivalents in a liter were as follows: NH_4^+ 1, NO_3^- 14; K^+ 6, H_2PO_4^- 1; Ca^{++} 8, SO_4^{--} 4; Mg^{++} 4.

Iron was added once or, as the plants became large, twice weekly at the rate of $\frac{2}{3}$ cc. of 0.5 per cent iron tartrate solution to a liter of nutrient solution.

The following grades of nutrient salts were used: KNO_3 and $\text{Ca}(\text{NO}_3)_2$ —fertilizer grade; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ —technical; and $\text{NH}_4\text{H}_2\text{PO}_4$ —C. P. The nitrate fertilizer salts were of synthetic origin. As already stated, the nutrient solutions were analyzed at frequent intervals, and a record was kept of the amounts of nutrients absorbed. Upon the depletion of a particular nutrient it was replenished in such a manner as to maintain, insofar as possible, the initial proportion of nutrient ions. As that was not always convenient or even feasible, the composition of the nutrient solution describes accurately the proportion of ions only at the beginning of the experiment.

The soil, of a clay-loam type, was similar to that used in a commercial greenhouse in the locality for the successful production of tomatoes. It was fertilized in a manner corresponding to its usual fertilization treatment under commercial conditions. In the fall-winter experiment 50 pounds per tank of well-rotted cow manure was mixed with the soil before sterilization for 6 to 7 hours in an autoclave. Five pounds of agricultural gypsum was added to the soil as it was being placed in the tank. At the conclusion of the first experiment the soil was sacked, autoclaved as before, and then returned to the original tank. The manuring treatment in the spring-summer experiment was increased to 100 pounds of well-rotted horse manure plus 50 pounds of well-rotted cow manure per tank; the gypsum was added as before. In the

⁴ We are indebted to W. A. Huelsen, of the Illinois Agricultural Experiment Station, for furnishing the seed.

spring-summer experiment, at the time of the heaviest setting of fruit, the soil was fertilized additionally with potassium and phosphate supplied in the distilled water.⁵

For the sand culture, included in the spring-summer experiment, pure white Belgian sand was used. A complete nutrient solution including iron and micronutrients was applied to the sand. A week after the plants were transplanted, a daily irrigation of 115 liters of nutrient solution was begun. The solution was allowed to drain freely, and no attempt was made to recover it.

Control of pH

The inclusion of a small proportion of ammonium nitrogen in the nutrient solution provided a very satisfactory "physiological buffer" in the sense suggested by Trelease and Trelease (15) and sufficed by itself to keep the pH of the nutrient solution within a physiologically tolerable range, fluctuating for the most part from pH 4.5 to 6.2. The ammonium nitrogen was added, as ammonium phosphate, whenever an analysis disclosed a need for replenishing the phosphorus. No attempt was made to maintain the ammonium concentration at any particular level other than that determined by the addition of ammonium phosphate. Thus, in this and in other experiments not included here, the pH of the nutrient solution took care of itself if the phosphate supply was replenished when needed, as $\text{NH}_4\text{H}_2\text{PO}_4$.

In the fall-winter experiment, however, phosphorus was added at certain times as KH_2PO_4 . Following these additions, several additions of H_2SO_4 were required to keep the reaction of the nutrient solution around pH 6.

Heating the nutrient solution

A previous report that heating the nutrient solution was associated with high yields (8) suggested the desirability of testing the effect of heating in all three nutrient media—soil, sand, and water culture—and comparing their respective yields when the same temperatures prevailed in the root zone. The lead heating cable and thermostat previously described (8) were used in all tanks, 120 feet of cable being placed in the soil and sand basins and 60 feet in the water culture tanks. The cable was painted twice with asphalt paint and, in the water culture tanks, was covered in addition with rubber tubing.

The temperature around the roots in the heated cultures was maintained day and night at $20^\circ \pm 1^\circ\text{C}$. in the fall-winter period, and at $23 \pm 1^\circ\text{C}$. in the spring-summer period. The temperature in the unheated soil and solution varied from 14° to 20°C ., depending on the air temperatures. The limitation in space prevented the inclusion of an unheated sand culture.

⁵ The applications of manure were large because of the dense spacing of plants. Similarly large applications of manure have been reported by two successful California growers of greenhouse tomatoes. To each soil tank, 2000 cc. of $M \text{KH}_2\text{PO}_4$ was added as insurance against any deficiency of potassium or phosphate during the heavy fruiting period.

The greenhouse itself was unheated except on a few occasions to prevent temperatures from falling below 10–13°C.

Aeration of the water culture

In previously reported large-scale water culture experiments with the tomato (8), no special provision was made for aeration other than lowering the level of the nutrient solution and using a porous litter cover over the tank which permitted gaseous diffusion. We have undertaken in the spring-summer experiment to investigate the effect of forced aeration, supplementary to that provided by the means described above, on the growth and yield of the plant and on its absorption of inorganic nutrients. Sintered glass aerators,⁶ which released compressed air in fine bubbles, were placed under each plant, making a total of twenty aerators in a tank.

Utilization of water

In the spring-summer experiment, records were kept of the amounts of water supplied to the soil and to the solution cultures. The level of nutrient solution in the tanks was maintained within several inches from the top of the tank, and the volume of water added each time was determined by means of a calibrated gauge attached to each tank. The usual commercial practice was followed in watering the soil.

Analytical procedures

All pH determinations were made by means of the glass electrode. Standard procedures were used in the analysis of the nutrient solution⁷ for calcium, magnesium, potassium, nitrate-nitrogen, and phosphate; the cobaltinitrite method was used for K; the phenoldisulfonic, for NO₃; and the colorimetric, for phosphate.

EXPERIMENTAL RESULTS

Comparison of yields from soil, sand, and water culture

One of the basic assumptions in some discussions of the commercial possibilities of artificial nutrient media has been that such media have inherent capacity for far higher yields than are attainable under even the most favorable soil conditions. This view has not been supported by direct comparisons with crops produced in soil but is based rather on comparisons of yields obtained in relatively small-scale water culture experiments (frequently in greenhouses) with average agricultural field yields for an entire state or even country. The

⁶ Presently, a simpler and more efficacious aeration technic was evolved. It consists of porous carbon tubes (available on the market) connected with rubber tubing to a compressed-air line and placed at the bottom through the entire length of the culture tanks. Air escapes in the form of very fine bubbles.

⁷ Hibbard, P. L. 1939 Methods of chemical analysis. Division of Plant Nutrition, University of California. Mimeographed.

reasons why such comparisons are unacceptable and why they may lead to mistaken inferences have been outlined elsewhere (9). Using the tomato crop as an example, we may remark here that field yields in California vary from 3 tons an acre for a spring shipping crop to 30 tons for canning tomatoes (13). Field yields are, of course, a function not solely of soil fertility but also of a host of other factors: variety, climate, irrigation, cultural practices, length of harvesting season, and damage by insects and diseases.

By using basins of the same area for each medium and by growing plants of the same variety in one climatic environment and with similar general cultural practices, we undertook to compare the yields of tomatoes from artificial nutrient media and from a fertile soil. It has been suggested that one advantage of water culture is its ability to support a denser stand of plants than a soil could support, and therefore by having the same number of plants per unit area in a soil as in a water culture one would limit the inherently higher

TABLE 1
Yields of greenhouse tomatoes grown in heated nutrient media
(Pounds of fruit harvested from an area of 25 square feet)

	FEBRUARY- AUGUST	12-MONTH PERIOD
Soil.....	238	358*
Sand.....	357	
Water culture.....	288	402*
Water culture, highest yield previously reported for similar conditions (7, p. 128).....		352

* These annual yields represent sums of the Crackerjack variety crop in the fall-winter season and the mixed Lloyd Forcing and Crackerjack crops in the spring-summer season.

productive capacity of water culture to that of the soil (7). We have selected, therefore, for all our basins the number of plants previously found to give a high yield in water culture (7, 8). As this spacing was considered justifiable only under good light conditions and was found too close for others (7), the inference may be drawn that it afforded full opportunity for comparing the supplying power for water and nutrients of soil and of artificial nutrient media.

The yields obtained are given in table 1. The highest yield by water culture previously reported for a similar area is included for comparison (7).

It is apparent that no evidence was found in support of the view that large-scale water culture technic (7, 8) has vastly superior crop-producing capacity to that of a fertile and well-managed soil. The significance of such differences in yield as were obtained between soil and water culture in our experiments cannot be established without further experimentation. The water culture yield of greenhouse tomatoes quoted from another source (table 1) was obtained with a variety of tomatoes different from that employed by us, but it may serve

here as an approximate guide in appraising the productivity of water culture in the hands of different workers. It offers no indication of extremely large increases of yields in water culture over those in a highly productive soil.

No provision for forced aeration was made in any of the water cultures considered in table 1. As subsequent discussion will disclose, forced aeration brought about a considerable increase in the yield of fruit from plants grown in water culture (table 3). The importance of the factor of aeration in evaluating the productive capacity of water culture versus soil will be considered later.

Our annual yields reported in table 1 are sums of fall-winter and spring-summer yields.⁸ The markedly higher spring-summer yields obtained in all media illustrate the dominating influence of light in greenhouse tomato production—a fact well established in commercial practice (6). The yields were obtained from two varieties grown in the same tank. Of these, Lloyd Forcing gave considerably higher yields than Crackerjack; had Lloyd Forcing been

TABLE 2

Effect of heating soil and nutrient solution on the yield of tomatoes at two seasons

TREATMENT	AVERAGE YIELD PER PLANT	
	Fall-winter, var. Crackerjack	Spring-summer, var. Lloyd Forcing
	lbs.	lbs.
Soil heated.....	6.0	14.2
Soil unheated.....	6.7	13.9
Solution heated.....	5.7	17.0
Solution unheated.....	5.5	15.7

grown exclusively, the yields per unit area would have been correspondingly higher.

Attention is drawn to the high yield of the sand culture cited in table 1. The yield from this tank for the 6-month spring-summer season compares favorably with yields for the entire year of the other cultures.

The general appearance of the plants in soil and in sand and solution cultures is shown in plate 1.

Effect of heating the nutrient solution and the soil

As shown in table 2, heating the nutrient solution or the soil in either the spring-summer or the fall-winter period produced no great effect on the yield of fruit. The mild and fairly uniform air temperatures prevailing the year

⁸ The water culture yield reported by Gericke (7) which was cited in table 1 for comparison with our yields was based on yields obtained from plants grown continuously for 12 months, with the major portion of the crop harvested in the first 8 months (8). Table 3 shows that our higher yielding variety (Lloyd Forcing) gave yields for 6 months comparable with those of the higher yielding variety (Sutton's Majestic) in the experiment referred to above.

around in Berkeley provided favorable greenhouse temperatures. It appears that if these are maintained there is no need for heating the solution. The average greenhouse air temperatures during most of the year were sufficiently high to maintain in the unheated cultures a temperature around the roots of about 18°C., which was adequate for growth. An interesting light is thrown on the adequacy of this temperature by the favorable influence of forced aeration in the unheated nutrient solution as shown in table 3.

Influence of aeration

The view has been expressed that by the use of suitably porous "seedbeds" and by allowing the level of the nutrient solution in the tanks to drop 2 inches, fully adequate aeration for the roots is insured and that although supplementary aeration may be needed when plants are grown in bottles kept filled with nutrient solutions or when growing conditions are otherwise unfavorable, yet

TABLE 3
Effect of aeration and heating on the yield of tomatoes (var. Lloyd Forcing) grown for 6 months from February to August

TREATMENT	AVERAGE YIELD PER PLANT	HIGHEST YIELD OF INDIVIDUAL PLANT
	<i>lbs.</i>	<i>lbs.</i>
Solution unheated.....	15.7	20.2
Solution unheated, aerated.....	21.1	28.0
Solution heated.....	17.0	24.1
Sand heated.....	21.6	32.4
Solution heated, yields previously reported (7, p. 128)...	16.2*	27.4*

* Annual yields, varieties Sutton's Best-of-All and Sutton's Majestic.

"when proper conditions are provided, bubbling air through the solution is unnecessary" (7).

On the other hand, it has been the experience of workers in this and other laboratories (5) that for many plants the oxygen supply normally available to the roots in a nutrient solution without forced aeration is suboptimal. That was true in a series of our experiments whether the level of the nutrient solution in a tank was allowed to drop or not. But as in all the experiments referred to, metal covers were employed with corks for supporting plants (9), we undertook to investigate the influence of forced aeration with a porous litter as a cover for the tanks.

Forced aeration was associated with a distinct increase in both vegetative vigor and fruitfulness. The yields are given in table 3. A comparison with the high yields reported by Gericke (7) is included in table 3 to indicate that plants grown in the unaerated cultures in our experiments were similarly fruitful. The level of the nutrient solution in the unaerated cultures was kept within about 2 inches of the top of the tanks.

Aeration gave an appreciable increase in yield over the unaerated cultures, whether heated or not. As previously mentioned, all plants were staked to extend to the full height of the greenhouse. Since fruit was borne over the entire length of vines, the highest yields were obtained from the tallest plants and lower yields from those which could extend only to the lower part of the sloping greenhouse roof. The highest yields from individual plants are given in table 3 to indicate the potentialities for fruit production with relatively unrestricted vertical extension of vines.

At the conclusion of the experiment, the roots from all the plants in a tank were excised, washed, and dried at about 75°C. The average dry weight of roots, in grams, per plant was recorded as follows: solution unheated, 12.4; solution unheated, aerated, 19.9; solution heated, 14.6. The favorable effect of aeration on root growth is thus illustrated.

The conclusion is drawn from these data that aeration was a limiting factor in the growth of tomato plants even when they were grown in tanks with porous covers and with a lowered level of solution. It is interesting to note that this favorable effect of aeration was obtained in the unheated cultures. Heating the nutrient solution gave but a small increase in yield. This suggests that oxygen supply rather than temperature was the dominant influence in determining the maximum growth and fruitfulness possible under the prevailing excellent light conditions.

Support for this conclusion is found in the evidence of increased absorption of inorganic nutrients, to be discussed presently, and in the excellent vegetative growth and yield of fruit obtained in the sand culture. It will be recalled that the sand was heated, and hence it can be compared to the heated soil—the ability of the latter to supply water and nutrients was well established. Attention has already been drawn to the higher yields in sand than in soil (table 1), and the yields from sand are comparable to those from the aerated water culture (table 3). The soil, as pointed out, was a clay loam and was well fertilized with manure. The excellent growth that the soil supported is *ipso facto* evidence that the soil did provide very good aeration for the roots. But the improved growth in the sand suggests that the plants could profit from a still more liberal oxygen supply, which was available in the sand culture. Possibly more rapid removal of CO₂ was also a factor.

Water economy of plants grown in soil and water culture

The view has sometimes been advanced that the water requirement of plants is smaller in water culture than in soil. Indeed some popular misconceptions that large-scale water culture holds out promise of producing crops in climatically favored desert regions seem to rest partly on this assumption. Obviously, even if crops produced by this method in desert regions required less water, the difficulties in providing a somewhat smaller water supply for water culture would frequently be essentially the same as those encountered in providing a larger amount of water for irrigation of soil. In any case, no

direct evidence was ever put forward that actually less water is required to produce a crop under similar climatic conditions in water culture than in soil.

The growing of tomatoes in soil and in water culture side by side in the same greenhouse afforded an opportunity to measure the relative amounts of water utilized. The numbers of gallons of water used to produce 100 pounds of fruit were as follows: soil heated, 214; soil unheated, 222; solution heated, 276; solution unheated, 257.

The results indicate that somewhat more water was used to produce a unit weight of fruit under water culture than under soil conditions. What seems to warrant emphasis, however, is not the difference, but the essentially similar general magnitude, of water utilization by the plants grown in either medium. This is in harmony with the view that the principal water loss of plants is through transpiration and is determined for the same species and climate by the total amount of transpiring surface. Because of the general similarity in growth of plants grown in soil and in water culture (pl. 1), it seems reasonable to assume that the amount of water transpired was of the same order of magnitude in both cases.

The method of adding water to the soil was not conducive to water loss by over-irrigation. Water was applied each time in such amounts as not to go beyond the chief zone of root development. Whether the advantage in water economy for the soil as shown in the foregoing figures is significant and whether it is to be attributed to less evaporation from soil than from the water culture surface cannot be evaluated on the basis of evidence now available.

In other experiments not reported here, a relation between aeration of the nutrient solution and water absorption by tomato plants was noted. After the plants attained a height of about 3 feet, wilting was observed on warm sunny days in the unaerated tanks. Recovery was brought about by bubbling air through the nutrient solution (this also accelerated removal of CO_2). Wilting was absent in plants grown in the same greenhouse in nutrient solutions with forced aeration and even in plants grown in unaerated solutions but in tanks covered with porous beds rather than with metal covers. Although the inclusion of a porous bed without forced aeration failed to provide optimum aeration conditions, yet it clearly provided better aeration than that which prevailed in the tanks with the metal covers. In this connection another possible factor is the development of roots in the aerated porous bed.

Absorption of nutrients from unheated, heated, and aerated nutrient solutions

The total amount of nutrients removed in 5 months by plants grown in the several nutrient solutions was computed from periodic analyses of the solutions and was expressed in terms of milliequivalents absorbed by a single plant (table 4). The period for which the data are given was from March 22 to August 24, when the experiment was terminated. The plants were germinated in sand irrigated with nutrient solution on December 10 and were transplanted into the respective tanks on February 14.

A marked increase in the absorption of all nutrients resulted from aerating the unheated solution and an intermediate increase from heating the nutrient solution. The data suggest that from the standpoint of mineral nutrition, aeration of the nutrient solution at the lower temperature was more important under our conditions than heating the solution. The fact that both aeration and heating increased the absorption of inorganic nutrients is in harmony with the general principle that active salt accumulation by roots is dependent upon metabolic activities of cells (10), although some passive intake and upward movement of salt under the influence of transpiration may take place through metabolically inactive or injured roots. Certain observations indicate, however, that with forced aeration, injurious effects on growth may be obtained from relatively high temperatures around the roots. These conditions lead to excessive metabolic activity.

TABLE 4
Absorption of nutrients in a 5-month period by tomato plants
(Milliequivalents per plant)

	UNHEATED NUTRIENT SOLUTION	HEATED NUTRIENT SOLUTION	UNHEATED, AERATED NUTRIENT SOLUTION
K ⁺	506.3	599.9	737.5
NO ₃ ⁻	775.9	853.7	1,074.2
H ₂ PO ₄ ⁻	117.5	142.7	160.0
Ca ⁺⁺	329.3	393.3	445.4
Mg ⁺⁺	140.7	142.5	197.2

Absorption of nutrients by tomato plants at various stages of development

The average daily absorption of nutrients from the aerated solution is graphically presented in figure 1. Potassium and nitrogen were absorbed in very high amounts throughout the season, but their absorption fell off distinctly in the last period. Calcium, magnesium, and phosphorus were absorbed in smaller amounts and their absorption was sustained throughout the season. The harvesting of fruit began on May 23, and the middle period in figure 1, from May 6 to July 7, coincided with the heaviest setting and the rapid development of fruit. The particularly prominent position occupied by potassium and nitrogen in the composition of both fruit and leaves of tomato plants accounts for the large demand for these nutrients throughout the season. On the other hand, the remaining three nutrients are not, on a quantitative basis, major inorganic components of tomato fruit but are quantitatively important constituents of the leaves. Sulfur was not included in these analyses. It is a nutrient whose concentration in the nutrient solution can undergo considerable variation without any important consequences to the plant.

It is difficult to interpret solely on the basis of the data here presented the significance of such seasonal fluctuations in absorption as are illustrated in figure 1. The plants in the aerated nutrient solution had excellent opportunity

for continued absorption of inorganic nutrients and may have exhibited "luxury absorption," as the nutrient solution was not permitted to become depleted. Inasmuch as the composition of the leaves in relation to the external supply of nutrients is a factor in the translocation of inorganic elements to the fruit (4), it is to be expected that stores of inorganic elements accumulated in the leaves at one period were transported to the developing fruit in another.

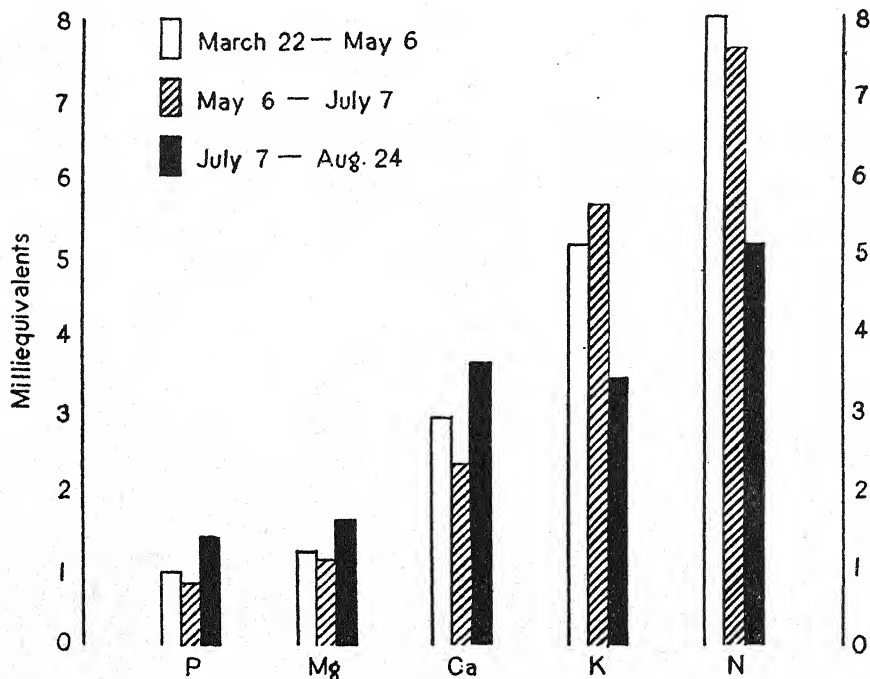


FIG. 1. AVERAGE DAILY ABSORPTION OF NUTRIENTS FROM AN AERATED SOLUTION BY A TOMATO PLANT AT VARIOUS STAGES OF GROWTH

The nutrients were assumed to have been absorbed in the ionic form as given in table 4

DISCUSSION

Is there an optimal nutrient solution?

Despite the early recognition by the originators of the water culture technic (14) that, within certain ranges of composition and total concentration, fairly wide latitude exists in the preparation of nutrient solutions suitable for plant growth, the question is sometimes asked whether there is some physiological basis for preferring one nutrient solution to another. Different concepts are sometimes suggested for arriving at an "ideal" nutrient solution, among them that the nutrient solution should reflect the "average" composition of the plant or that there should be a "balance" between the various nutrients.

Before discussing the bearing that the results of this investigation have on

the selection and management of nutrient solutions, certain general considerations may be reviewed. It is apparent that in nature plants do not grow in nutrient media of fixed composition. Indeed the opposite is the case, for not only do plants grow in soils widely differing in their power of supplying nutrients, but also there occur in any one soil wide seasonal fluctuations in the nutrients that were available at the beginning of the growing season. The crucial factor from the standpoint of the optimal development of the plant is the availability and accessibility at each stage of growth of a *sufficient quantity* of each essential element, within suitable total concentration ranges and fairly broad limits of ionic proportions.

The composition of a plant cannot offer a universal guide in devising nutrient solutions. Plant composition is in part genetically determined and is influenced by climatic factors. Different kinds of plants grown at the same time in the same soil or nutrient solution will develop tissues with different proportions of inorganic constituents. Moreover, the inorganic composition of various parts of a plant may vary considerably and, to use tomato as an example, a different nutrient solution may be suggested by the composition of the fruit than by the composition of the foliage. Also the influence of climatic factors on the utilization of inorganic elements, particularly nitrogen, needs to be taken into account.

In addition to the considerations advanced above, it is clear that since the composition of the nutrient solution affects, within certain limits, the composition of the plant, it is not logical to assume that an analysis of the plant affords an accurate basis for formulating a nutrient solution of unique physiological properties.

An analogy with soil conditions offers no compelling reason for either selecting or maintaining an originally selected "best" nutrient solution. The experience of a host of workers in numerous laboratories throughout the world with a number of different nutrient solutions supports this view. In a strict sense, because of the differential absorption of nutrients by the plant, no one nutrient solution retains its identity after the plants have grown in it for some time, unless large volumes relative to the number of plants are made available, either by means of a continuously replenished "flowing" solution or by growing relatively few plants in tanks filled with large volumes of nutrient solution.

Within relatively wide limits of osmotic concentration, of pH values, and of concentrations of individual salts, the problem of selecting a good nutrient solution is chiefly one of selecting a solution *most conveniently* managed to insure an adequate supply of nutrients. This is illustrated by the following. In the present investigation the aerated nutrient solution produced tomato plants of excellent vegetative growth and fruitfulness. It can be assumed, therefore, that the absorption of nutrients from this solution, as shown in figure 1, represented an adequate balance between the requirements of the plant and the supply of nutrients in the solution. As already mentioned, the nutrient solution was analyzed from time to time, and the absorbed ions were replen-

ished. It is obvious that the elements absorbed in the largest amounts, potassium and nitrogen, were replenished the oftenest. An alternative course would have been the discarding of the entire solution when one or more nutrients became depleted and replacing it with a new solution.

Changing the entire solution or replenishing individual nutrients offers a practicable way of managing nutrient solutions. Much would be gained, however, if the original composition of the nutrient solution were to approach the actual proportions in which the several nutrients were absorbed from the solution. This would allow for a simultaneous depletion of all the nutrients and would render it unnecessary to add, upon frequent analysis, individual salts. Instead, a new supply of all the nutrient salts could be added to the solution when the original supply approached exhaustion. The farther the original composition of the nutrient solution departs from the actual proportion of absorbed ions, the less advisable would this relatively simple procedure be, because of the accumulation of residues of ions absorbed at slower rates.

TABLE 5

Proportions of ions in original solution compared with those absorbed by the plant from the aerated nutrient solution during a 5-month period

	ORIGINAL NUTRIENT SOLUTION	ABSORBED BY PLANT
	<i>m.e.</i>	<i>m.e.</i>
NO ₃ ⁻	14	14
K ⁺	6	9.1
Ca ⁺⁺	8	5.8
Mg ⁺⁺	4	2.6
H ₂ PO ₄ ⁻	1	2.1

In table 5 a comparison is made between the proportions of nutrients in the original solution and those computed from amounts of the various ions actually absorbed by the plants from the aerated nutrient solution (table 4). The proportions of absorbed ions as given in table 5 and figure 1 represent averages over appreciable periods of time and cannot be taken as defining the daily absorption of nutrients. They are regarded rather as approximate guides in tracing the uptake of nutrients by tomato plants.

More potassium and phosphorus in relation to nitrogen was absorbed than corresponded to the proportions in the nutrient solution. Reducing the calcium nitrate and increasing the potassium nitrate, as well as doubling the phosphorus, would render the proportions of the ions in the nutrient solution more comparable with the proportions of nutrients actually removed by the plants. The higher proportion in the nutrient solution of magnesium with its accompanying sulfate ion is of no great consequence and could be left unchanged.

Another problem in the management of nutrient solutions arising from the differential absorption of ions is that of hydrogen-ion concentration. The

net result of a preponderance of anion over cation absorption is a decrease in hydrogen-ion concentration which brings in its train, at neutral or alkaline reaction, the precipitation of such nutrients as iron, manganese, and phosphate. The shift toward neutrality or alkalinity is the usual course of events when nitrate salts are used as the sole source of nitrogen. On the other hand, a preponderance of cation over anion absorption results in an increase in hydrogen-ion concentration. This is usually encountered only when appreciable amounts of NH_4^+ are present in the nutrient solution. Otherwise, unless NO_3^- should be temporarily depleted in the presence of considerable K^+ , the problem of progressively increasing acidity of the nutrient solution will seldom arise. It is desirable to have at all times a slightly acid reaction of the nutrient solution, to avoid problems of precipitation, but this by no means implies that it is necessary to maintain a constant pH. Our experience with many species of plants has been that they grow very well in solutions ranging in pH from 4.5 to 6.5. As mentioned before, this fluctuation in pH occurred in the nutrient solution used here because a small portion of its nitrogen was added in the form of NH_4^+ .

We were guided by these various considerations in devising a "convenient" solution to meet the following requirements: (a) its proportion of nutrients should approach the actual removal of ions by the plants; (b) its pH should remain within a range favorable for plant growth and should require little attention between changes or replenishments of the nutrient solution. On the basis of the present data a solution of the following composition was prepared: KNO_3 , 0.010 M; $\text{Ca}(\text{NO}_3)_2$, 0.003 M; MgSO_4 , 0.002 M; and $\text{NH}_4\text{H}_2\text{PO}_4$, 0.002 M. In preliminary trials this solution was tested with tomatoes and lettuce. The pH of the solution required no attention, and the uptake of minerals was conducive to excellent growth.

It is evident that this solution does not furnish nutrients in the exact proportions in which they are absorbed at every stage of the growth of the tomato plant (fig. 1). Nevertheless, it was found to be a very favorable nutrient medium for the growth of tomatoes and economical from the point of view of convenience in management. This solution was also found to be favorable for the growth of various other kinds of plants. Yet it may be necessary to alter its composition to accord with the different capacities for absorbing the several nutrients, characteristic of each kind of plant, if it is desired to approximate in the solution the proportions of nutrients actually absorbed. Of course, even for the same kind of plant, absorption of nutrients from the same nutrient solution will vary in accordance with the rate and type of growth as influenced by climatic and seasonal conditions.

Nothing has been said, so far, about the proportions of micronutrients. Obviously as they are required in exceedingly small amounts, they do not influence by mass effects the ionic relations discussed above. For many plants, zinc, copper, and molybdenum may not have to be deliberately added, as they will often be adequately supplied as contaminants in the nutrient

medium. Boron and manganese are generally added to culture solutions to give a concentration of 0.5 p.p.m. each.

As pointed out earlier, the aeration requirement of plants is variable, and for many species, bubbling air through the solution may be a highly desirable, if not an indispensable practice. The amount of air normally gaining access to the solution may be inadequate for some plants even when a porous cover is used and the level of the solution in the tank is allowed to drop. The importance of aeration in nutrient solutions needs stressing. In many instances, plants may fail because of inadequate aeration in an otherwise well-managed nutrient solution.

Supply versus concentration of nutrients

The question is sometimes asked whether the entire requirement of nutrient salts needed to carry plants to maturity may be initially incorporated in the nutrient solution without producing injury from excessive osmotic concentration. Theoretically that is possible if no restriction is placed on the volume of the nutrient solution in relation to the number of plants. It is seen from table 4 that the aggregate absorption of K^+ , NO_3^- , $H_2PO_4^-$, Ca^{++} , and Mg^{++} by one tomato plant for a period of 5 months was 2614.3 m.e. Since 1 liter of nutrient solution provided 33 m.e. of these ions (disregarding for the present purpose the difference in proportions), it follows that about 80 liters of nutrient solution would have been required to supply the needs of one plant for 5 months. Our tanks provided for about 20 liters of solution to a plant, and consequently it was necessary to replenish the solution from time to time. By using larger tanks or by reducing the number of plants, less frequent replenishments of nutrient solution can be made to suffice.

Some latitude is permissible in increasing the concentration of the nutrient solution before ill effects are brought about. In one experiment good vegetative growth and fruitfulness was obtained with tomatoes grown in a concentration 2.5 times the one used here. But plants differ in their salt tolerance, and no generalizations seem warranted. Also the question of climatic influences, particularly that of light in relation to nitrogen concentration needs to be considered in this connection (12).

Most nutrient solutions have total concentrations of salts equivalent to osmotic pressures ranging from 0.5 to 1 atmosphere. This range seems to be well suited to the physiological tolerances of most plants.

Large- versus small-scale water culture

No evidence was found in support of the view that large-scale water culture operations rest on new principles unknown to the usual small-scale scientific experimentation. Such departures from the small-scale technic as are observed in crop production in water culture are determined in every instance by the scale of operations and by economic considerations. It is evident that, depending upon the particular object of a scientific experiment in which the

water culture method is used, more or less rigid attention has to be given to the selection of containers, chemicals, and water. On the other hand, if the sole objective is to produce crops in water culture, the cheapest material suitable for the purpose may be selected. Thus ordinary tap or irrigation water of good quality can replace distilled water, and fertilizer grade salts, if free from toxic impurities, can replace the pure chemicals used by the scientific investigator. The ever-increasing purity of commercial fertilizers makes their use entirely possible at present. In point of fact, the commercial grades of calcium and potassium nitrates, of synthetic origin, were found in this laboratory to contain smaller quantities of impurities of heavy metals than certain C. P. grades. Likewise, any sturdy and water-tight material, if it is not toxic to plants, may be used for the construction of basins. Other matters of technic, such as the method of supporting the plants and the manner of adding nutrients to the solution, are determined chiefly by consideration of convenience.

Since the use of water culture instead of soil in producing crops does not alter climatic requirements, obviously these must be fulfilled to bring about optimum growth. Light-temperature relations and length of day must be carefully considered for each crop and for the locality in which it is to be grown. The fluctuation in the temperature of the nutrient solution is but one consideration and should be evaluated in relation to the other climatic factors.

Possibilities of large-scale nutrient solution technics

The results of our experiments confirm earlier views that the possibility exists of producing crops on a large scale by the water culture or the sand culture method. By careful management of artificial nutrient solutions with respect to availability of all nutrients and by attention to optimum aeration requirements of different plants, excellent yields can be produced which would compare favorably with those obtained from the best soils under comparable climatic conditions. But as pointed out previously (3, 9), the application of the water culture method for crop production will be determined by economic considerations which would limit the use of the method primarily to high-priced greenhouse crops or to unusual situations in which good soil is not available (for example, coral islands). Obviously, field crops can be produced more cheaply by conventional agricultural methods, even though higher yields could be obtained in favorable nutrient solutions than in soils.

It has been suggested (7) that one advantage of water culture is its ability to support multiple or mixed cropping in the same area, chiefly because the nutrient solution can be easily supplied with enough nutrients for several crops. But again the successful growth of dense vegetation by multiple cropping will depend primarily on the availability of light in a given area. Assuming that light is not limiting and that problems of shading are overcome, it remains to be seen whether by liberal fertilization and irrigation some soils cannot support as dense a stand of vegetation as a well-managed nutrient

solution. Whether the practice of multiple cropping could be recommended for either soil or large-scale water culture would depend chiefly on economic and climatic considerations. Too close planting or mixed cropping might render the use of labor-saving machinery in planting, harvesting, etc., infeasible and consequently increase the cost of production.

General comments on factors limiting yields

The comparisons of solution, sand, and soil cultures lead to consideration of several general questions of limiting factors for plant growth under natural conditions. First, a distinction should be made between limitation of yields imposed by climatic factors and by factors traceable to the nutrient medium; at the same time the interrelations between climatic and nutrient conditions must be recognized. Obviously, when climate is *per se* a limiting factor, no improvement in an otherwise well-adjusted nutrient medium, be it soil or water culture, can bring about an increase in yields. The well-known dominant influence of light on the yield of greenhouse tomatoes as illustrated in table 2 is a case in point. As in commercial practice (6), we found that with the same nutrient medium and spacing of plants the spring yields of greenhouse tomatoes are usually more than twice as high as the fall-winter yields. Aside from such limited favorable adjustments as can be made in nitrogen supply under conditions of relatively deficient light, no manipulation of the nutrient medium can, of course, compensate *in toto* for a deficiency of light.

As for the influence of the nutrient medium itself on growth and yields, we have already noted that our aerated (with forced aeration) culture solution as well as the sand culture gave a considerably higher yield of fruit than did the soil employed in that particular experiment. Subsequent experience with several other soils gives reason to suppose that the soil in question was exceptionally favorable for the growth of tomato plants. It had been heavily treated with manure and chemical fertilizers, and it must have had a high supplying power for essential nutrients. But it seems doubtful if even the best soil supplied oxygen to the roots at as high a rate as the culture solution through which rapid streams of air were passed. In addition to the rate of oxygen supply, there is the factor of CO₂ removal, and full differentiation of the two factors cannot be made on the basis of present evidence. We believe that our experiments suggest the possibility that in a well-fertilized soil, aeration (or associated CO₂ environment) may often limit the growth of certain crops. Aeration conditions in soils will be of particular consequence to crops which, like tomato, have a high aeration requirement. The importance of aeration in fertile soils for other crops which, like cereals, are not noted for their high aeration requirements, would seem to be relatively less important. That aeration can be a limiting factor in nutrient solutions has already been shown (table 3). But the improvement of aeration in artificial nutrient media as contrasted with soil is more readily accomplished, and herein may lie the inherent advantage of such media over soil, as far as the growth of certain kinds of plants is concerned.

The vigorous growth and the high yields of the tomato plants in the inorganic solutions imply that organic matter *per se* in the soil medium did not serve any indispensable purpose. Evidently the plants were capable of synthesizing their requirements of growth substances, as well as those of other organic constituents. In recent years the idea has been advanced that some types of plants will respond to the addition to the soil or other culture medium of vitamin B₁ or perhaps other organic substances. Experiments with inorganic nutrient solutions made in this laboratory (2) so far do not confirm this view, even when species of plants have been tested for which such responses have been claimed. Obviously organic matter present in a soil may have many very important secondary effects, but the direct need of organic constituents in the nutrient medium for the growth of higher plants is not satisfactorily demonstrated.

The artificial culture solutions in which the tomato plants were grown had as an initial composition much higher concentrations of several nutrients, particularly potassium and phosphate, than most soil solutions. The interpretation of culture solution data in terms of the soil is made more difficult because of the contact effects between roots and soil colloids—a subject which has been discussed during the last few years (11). Since, however, nearly all the nitrate ions, and equivalent amounts of cations, are present in the soil solution, the latter must be an important immediate source of nutrients in soils. But potassium and phosphate ions may be so low in concentration in some soil solutions as to raise the question of direct movement of adsorbed ions from the soil colloid into the root by ionic exchange. On the other hand, there are indications from soils thus far investigated that when the soil has adequate power to supply enough potassium to enable good plant growth, the concentration of this nutrient in the soil solution is not of a lower order of magnitude than that found to be sufficient (in flowing nutrient solutions) for the rapid growth of tomato and other plants, at least in the vegetative stage. Concentrations of phosphate in displaced soil solutions may sometimes be so low that the absorption of phosphate by the plant cannot be accounted for by examination of the displaced solution.

So far as we are aware, it has not been shown that under greenhouse conditions a heavily fruiting plant can be developed with extremely low concentrations of potassium or phosphate either in an artificial nutrient solution or in a soil solution. In our experiments, the soil solution concentrations of these nutrients were relatively high as a result of the treatments given to the soil.

It is necessary to keep in mind the two factors of concentration (including degree of saturation of soil colloid) and of the total supply in both soils and artificial nutrient solutions. In the soil, as in the solution, the volume of the medium accessible to the roots should not be disregarded. For example, in certain cropping experiments with a soil of low potassium availability, the appearance of potassium deficiency symptoms within a limited time depended on the size of the container in which the plants were grown.

An interesting observation was made on the relation of nitrogen supply to vegetative growth and fruitfulness in tomatoes grown under our conditions. In the spring-summer season all plants made excellent vegetative growth and were also highly fruitful despite the very large supplies of nitrogen in the nutrient medium, that is, supplies probably sufficient to enable the plants to absorb nitrogen to their full capacity for absorption. This was particularly true in the sand culture where, because of the daily irrigation with nutrient solution, the problem of the depletion of the nutrient solution with time did not arise. It seemed that the well-known relation between high nitrogen supply and dominantly vegetative growth habit in tomatoes [cf. review (12)] did not operate under our conditions. This may be attributed to the highly favorable light-temperature relations during the day and to the relatively cool nights conducive to conservation of carbohydrates, owing to low respiration.

SUMMARY

Experiments are reported on tomato plants grown under comparable conditions in soil and in sand and water culture media. Data on yields and absorption of nutrients are analyzed. The inherent capacity for crop production of a highly productive soil and of sand and water culture media was found to be of the same order of magnitude. Media affording liberal aeration (sand culture and water culture with forced aeration), however, gave higher crop yields than did soil. The suggestion is considered that aeration may limit crop production in some soils, even when the supply of water and nutrients is adequate.

Under the climatic and greenhouse conditions of the experiments, heating the nutrient medium did not lead to definite increase in rate of crop growth or in yield of fruit.

The use of water by the crop was of essentially the same magnitude for both soil and water culture conditions.

The complex question of the physiological basis for determining the composition of nutrient solutions is discussed. A convenient solution was devised based on the average amounts of nutrients absorbed by tomato plants in the water culture experiment.

A brief general analysis is attempted of certain limiting factors in crop growth as indicated by the experimental evidence obtained.

Previous opinion is reaffirmed that the use of the water culture method for commercial purposes depends on economic considerations, not on fundamental differences between soil and water culture media, or between small- and large-scale water culture methods.

REFERENCES

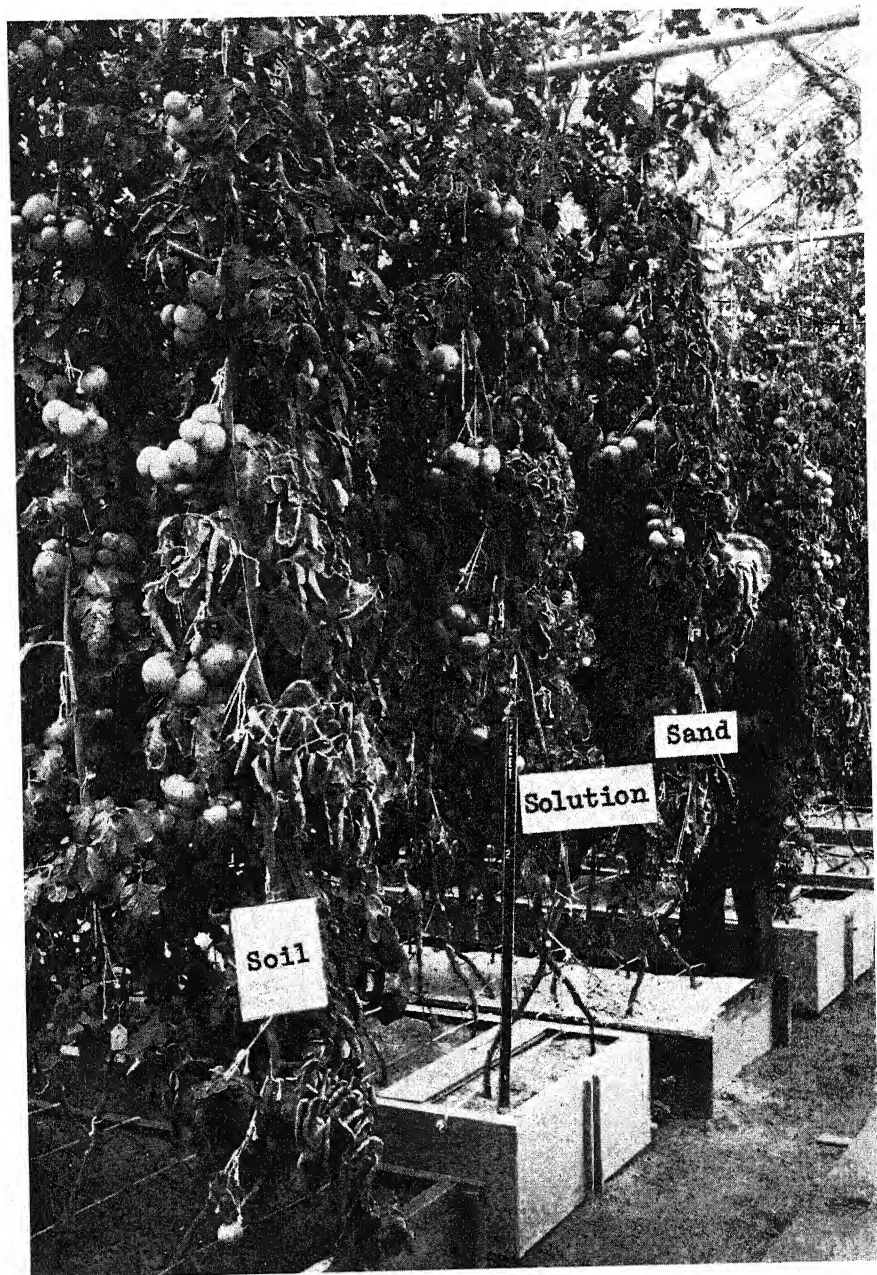
- (1) ARNON, D. I. 1938 Microelements in culture-solution experiments with higher plants. *Amer. Jour. Bot.* 25: 322-325.
- (2) ARNON, D. I. 1940 Vitamin B₁ in relation to the growth of green plants. *Science* 92: 264-266.

- (3) ARNON, D. I., AND HOAGLAND, D. R. 1939 A comparison of water culture and soil as media for crop production. *Science* 89: 512-514.
- (4) ARNON, D. I., STOUT, P. R., AND SIPOS, F. Radioactive phosphorus as an indicator of phosphorus absorption of tomato fruits at various stages of development. *Amer. Jour. Bot.* In press.
- (5) ARRINGTON, L. B., AND SHIVE, J. W. 1936 Oxygen and carbon dioxide content of culture solutions in relation to cation and anion nitrogen absorption by tomato plants. *Soil Sci.* 42: 341-357.
- (6) BEATTIE, J. H. 1939 Greenhouse tomatoes. *U. S. Dept. Agr. Farmers' Bul.* 1431: 1-28.
- (7) GERICKE, W. F. 1940 The Complete Guide to Soilless Gardening. Prentice-Hall, New York.
- (8) GERICKE, W. F., AND TAVERNETTI, J. R. 1936 Heating of liquid culture media for tomato production. *Agr. Engin.* 17: 141-143.
- (9) HOAGLAND, D. R., AND ARNON, D. I. 1938 The water-culture method for growing plants without soil. *Calif. Agr. Exp. Sta. Cir.* 347: 1-39.
- (10) HOAGLAND, D. R., AND BROYER, T. C. 1936 General nature of the process of salt accumulation by roots with description of experimental methods. *Plant Physiol.* 11: 471-507.
- (11) JENNY, H., AND OVERSTREET, R. 1939 Cation interchange between plant roots and soil colloids. *Soil Sci.* 47: 257-272.
- (12) NIGHTINGALE, G. T. 1937 The nitrogen nutrition of green plants. *Bot. Rev.* 3: 85-305.
- (13) PORTER, D. R., AND MACGILLIVRAY, J. H. 1937 The production of tomatoes in California. *Calif. Agr. Ext. Serv. Cir.* 104: 1-61.
- (14) SACHS, J. VON 1887 Lectures on the Physiology of Plants. Clarendon Press, Oxford.
- (15) TRELEASE, H. M., AND TRELEASE, S. F. 1935 Changes in hydrogen-ion concentration of culture solution containing nitrate and ammonium nitrogen. *Amer. Jour. Bot.* 22: 520-542.

PLATE 1

GROWTH OF TOMATO PLANTS IN FERTILE SOIL, IN NUTRIENT SOLUTION, AND IN
PURE SILICA SAND IRRIGATED EACH DAY WITH NUTRIENT SOLUTION

Fruit had been harvested for 7 weeks prior to photographing. All plants made excellent growth and set large quantities of fruit in all three media. The general cultural conditions—spacing, staking, etc.—were the same (Reproduced from *Calif. Agr. Exp. Sta. Cir.* 347).



BOOKS

An Agricultural Testament. By SIR ALBERT HOWARD. Oxford University Press, New York, 1940. Pp. 253, illus. 24. Price \$4.50.

The most interesting and suggestive book on soil fertility which has appeared since King's *Farmers of Forty Centuries*. Its thesis is that the natural organic manures, and the composts which are made by using them in conjunction with other farm wastes, are far superior to the artificial inorganic fertilizers. The Indore Process of manufacturing such composts is considered in detail. The mycorrhizal association between the soil humus and the roots of plants is credited with effecting such improvement in the well-being of the plants as to enable them to ward off the attacks of diseases.

Annual Review of Biochemistry. Volume IX. Edited by JAMES MURRAY LUCK AND JAMES H. C. SMITH. Annual Reviews, Inc., Stanford University, 1940. Pp. 744. Price, \$5.

This annual review of the progress of the biochemical sciences fills a highly important place on the shelves of the workers in this field. The present volume covers 26 subjects, including chapters on oxidations and reductions; the chemistry of glucosides, acrylic constituents of fats and oils, lipins, sterols, and amino acids; the metabolism of sulfur compounds, fats, carbohydrates, and proteins; discussions of enzymes, hormones, vitamins, malignant tissues, plant pigments, viruses; and reviews of such diverse topics as inorganic metabolism and organic acids of plants, microbiology of soils, biochemistry of fungi and insects, microchemical analysis, and radioactive indicators. Over 4,000 references are appended to the several chapters.

Better Lawns. By HOWARD B. SPRAGUE. McGraw-Hill Book Company, Inc., New York, 1940. Pp. 205, illus. 35. Price, \$2.

That the science of grass growing is replacing the art is well illustrated in this book, which deals with the turf problems on lawns and in parks. The information contained in the book is based on the results of many years of experimental work carried on at the New Jersey College of Agriculture under the author's direction.

Calcium Superphosphate and Compound Fertilizers: Their Chemistry and Manufacture. By P. PARRISH AND A. OGILVIE. Chemical Publishing Co., Inc., New York, 1939. Pp. 322, illus. 149. Price, \$14.

A thorough revision of a standard English work, formerly entitled *Artificial Fertilizers: Their Chemistry, Manufacture and Application*. The book deals

primarily with modern methods of manufacture of superphosphates but includes a special chapter on the crystallization and granulation of fertilizer materials. It is of interest to note that the authors raise the question whether solubility in water, the only test applied to European superphosphates, is a true guide to the availability of the phosphoric acid that is contained in them.

French-English Science Dictionary. By LOUIS DE VRIES. McGraw-Hill Book Company, Inc., New York, 1940. Pp. 546. Price, \$3.50.

This 43,000-word dictionary is the first of its kind to include words and phrases which are in common use in the agricultural sciences. The volume is compact in size, easy to use, and yields a quick answer to the questions which one ordinarily puts to a language dictionary. The buyer of this book will not be disappointed in his purchase.

General Bacteriology. By D. B. SWINGLE. D. Van Nostrand Company, Inc., New York, 1940. Pp. 313, illus. 157. Price, \$3.

A well-arranged presentation of the fundamental principles of bacteriological science and its application in industry and in the control of disease. The book is interesting and easy to read, yet it contains all the essential facts necessary to a thorough understanding of this subject. The book is very attractively bound.

The Home Gardening Encyclopedia. Second Edition. By WALTER BRETT. Chemical Publishing Co., Inc., New York, 1940. Pp. 448, illus. 19. Price, \$2.50.

A reference book for growers of fruits, flowers, and vegetables as a home-garden enterprise. Almost every operation which any gardener is called upon to perform, from the planting of abelia to the pruning of clematis, the paving of a pergola, the making of a rock garden, and the propagation of Zygopetalum, is fully described in this interesting book.

Insect Transmission of Plant Diseases. By JULIAN G. LEACH. McGraw-Hill Book Company, Inc., New York, 1940. Pp. 615, illus. 238. Price, \$6.

Introduced as an ornamental plant, the prickly pear (*Opuntia*) became a pest in Australia and spread over 60 million acres of land. To check its spread, the Land Commission imported insect enemies of the cactus from Texas and the Argentine. These insects, and the microorganisms which they transmit, have been so effective that 25 million acres of the land have been reclaimed. This is one of many highly interesting examples of insect transmission of plant diseases which are commented on in this pioneering text book in its field.

Organic Reagents Used in Quantitative Inorganic Analysis. By WILHELM PRODINGER. Translated and amended from the second German edition

by Stewart Holmes. Nordeman Publishing Company, Inc., New York, 1940. Pp. 203. Price, \$5.

An exceptionally useful reference work for those who are interested in analytical procedures as applied to the quantitative determination of trace elements in soils and plants.

Soil Science Society of America Proceedings. Volume 4. The Soil Science Society of America, Morgantown, West Virginia, 1940. Pp. 455. Price, \$5.

This large volume contains 89 complete papers presented at the meetings of the Society in New Orleans, Louisiana, in November, 1939, and abstracts of 38 other papers presented at a joint session with Commission III, International Soil Science Society, in New Brunswick, New Jersey, in August, 1939. The papers deal with such diverse soil subjects as aggregation, permeability, temperature, moisture, base-exchange, phosphorus, organic matter, boron, cotton fertilizers and diseases, forests, genesis and morphology, erosion, modification of soils due to human activities, legume bacteria, azotobacter, and biological processes. Considered in conjunction with the three preceding volumes, it presents a running account of the activities of the soil scientists of America. One cannot but be impressed with the great variety of applications of the fundamental sciences which are being made to this important field of endeavor.

THE EDITORS

Textbook of General Horticulture. By JULIAN C. SCHILLETTER AND HARRY W. RITCHEY. McGraw-Hill Book Company, Inc., New York, 1940. Pp. 367, illus. 136. Price, \$3.

A survey of the whole field of horticulture, including fruits, vegetables, and ornamentals, and discussions of problems involved in their production. At the end of each chapter are questions designed to test the student's knowledge of the subject and to stimulate his curiosity as to the "whys" of certain horticultural practices.



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